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# A MILKWEED SURVEY IN ONTARIO AND ADJACENT QUEBEC[1]

H. GROH[2] AND W. G. DORE[3]

*Science Service, Ottawa, Canada*

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Revival of interest in common milkweed (*Asclepias syriaca* L.) as a possible source of substitutes for products curtailed by the war was the occasion for the present survey activity.

The Dominion Government in 1942 instituted a search for native sources of rubber and other materials needed in the existing situation and, finding milkweed a promising species, proceeded to secure a quantity of the seed through the agency of co-operating schools in Carleton County adjacent to Ottawa. In 1943 land in Peterborough County, as well as at Ottawa, was seeded to milkweed; and a campaign for the harvesting of "wild" leaves for use in a pilot plant at Ottawa, experimentally producing milkweed "rubber," secured its immediate needs from collectors all over the province and beyond. In 1944 the demand was altered to pods, desired for their floss, to meet urgent requirements of a good substitute for kapok in providing life jackets for sailors and airmen. All the foregoing presupposed that milkweed in some quantity was, or could be made, available.

The general distribution and incidence of the plant was already known from data accumulated in the files of the Canadian Weed Survey over a period of some twenty years. This information, together with what could be gathered from various Canadian herbaria, has been published (1) but may be reviewed briefly here.

Common milkweed was found to be growing in all provinces from Saskatchewan eastward with the exception of Prince Edward Island and, until fairly recently, Nova Scotia. Its distribution westward from Ontario is interrupted north of the upper Great Lakes but not directly westward through Michigan; and the United States range continues southward even, according to some, to Florida, Texas and Arizona. Through much of this broad extent it is not sufficiently abundant for large scale exploitation. In Canada, east of Quebec city, west of Lake Huron, and north of Old Ontario the incidence is not high, occurrence being only in scattered patches, thin stands, or more often northward, not at all.

It was, then, within the southern portions of Quebec and Ontario that a more detailed survey, particularly for density, was required. Certain regions of higher incidence along an axis across Ontario had been indicated,

[1] Contribution No. 808 from the Division of Botany and Plant Pathology, Science Service, Department of Agriculture, Ottawa, Canada.
[2] Associate Botanist.
[3] Seasonal Agricultural Assistant.

but fuller and more quantitative data were still needed for intelligent direction of campaign effort as well as for the best possible understanding of habitat and other growth requirements of the plant. During 1943 and 1944 the available time of the joint authors was used in an attempt to spread as uniform and adequate a network of survey lines over Old Ontario as was possible under current transportation limitations. This was extended northward and eastward into Quebec, but not sufficiently to warrant inclusion here except for some portions along the Ottawa River.

## THE SURVEY

In any survey of such a large area some sort of random sampling method must be used. To this end efforts were made to traverse as many townships, or at least counties, as possible. Travel was by train or bus and by automobile. Auto travel allowed scrutiny of both sides of the road and the adjacent fields. By train one side only was recorded regularly although general impressions of fields opposite were gathered. Speed of some trains detracted from, but seldom wholly prevented reasonably satisfactory work.

Milkweed, being a tall and conspicuous plant (Figures 1 and 2) was amenable as many others would not be to this type of running survey. The species with which it was most likely to be confused at a distance were such as elecampane and low shoots of choke cherry. Concentration on a single object of survey permitted attention to density which was not possible in the earlier surveys covering hundreds of weed species at once for mere listing. In those circumstances an idea of incidence, derived from computation of the percentage of surveys in which milkweed appeared, was the primary objective.

The routes followed in the course of the Ontario surveys during the two summers are shown in Figure 6. Observations were made over a total of 5,860 miles, travelling 4,598 by automobile and 1,262 by train or bus. In Quebec adjacent to the Ottawa River 272 miles were travelled by automobile. It is clear that the survey even now is not exhaustive although distance from a line of travel is nowhere very great. Parts crossed in reaching others more remote were inevitably worked the more closely as, for instance, Nepean Township next to headquarters, in which scarcely a road was left untravelled. This resulted in data based on uneven amount but still quite comparable character of survey. Whatever selection there was of routes for auto travel aimed to use both primary and secondary roads and to reach the various parts of a municipality. Roads along political boundaries were avoided since estimates here required extra calculation to share them fairly between contiguous townships.

### *Method of Estimation*

In actual practice methods were evolved for separate recording from two distinct types of site: (a) along roadsides and railways stands of varying density, on belts from road shoulder to fence strip, of varying width especially when reduced by shrubbery, water or rock exposure, were more or less continuous and lent themselves to linear rather than area recording as they passed under view; (b) field stands within easy range

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FIGURE 1. Milkweed in flower is an attractive and distinctive plant. Of the numerous flowers going to make an umbel only a few mature as pods. These plants grew on the soil of an upland outcrop of crystalline limestone in prevailingly Laurentian gneiss at Lascelles, Gatineau Co., Quebec. (Photo by E. G. Anderson).

of vision beyond fence strips tended to be discontinuous owing to differences of land utilization in successive fields, woodland cover or drainage, and when present in enough area and density were recorded on an area basis.

Concepts of density first had to be established. Five grades from practically "absent" to "good" were found of practical value for use at a moderate rate of travel. Numerical equivalents for each of these were arrived at by counts of numerous stands that were considered representa-

FIGURE 2. Roadside plants are often vigorous and highly productive. These dust-covered plants bear respectively 17, 21 and 14 developing pods per stalk. This is a region of clay soil, 1½ miles east of Stanley Corners, Carleton County. The plants are growing along the shoulder of a dirt road next to a deep ditch. Other plants have been grazed down. (July 31, 1944).

tive. These are shown in Table 1 and in part illustrated in Figures 3, 4 and 5. For roadsides these were obtained by counts of all stalks in a roadside strip measured as to length, but merely average as to breadth. For field stands the count was of all stalks in a belt transect paced 70 yards long and 1 yard wide by sight, or approximately 1/70th acre in area.

Usually several transects in representative positions in a field were averaged to get a fair equivalent for the density given. The numerical equivalents so derived were used in all subsequent calculations.

TABLE 1.—SCHEDULE OF GRADES USED IN THE ESTIMATION OF DENSITY OF ROADSIDE AND FIELD STANDS WITH THEIR EQUIVALENTS IN NUMBER OF INDIVIDUAL STALKS

| Density grades | Roadside stands (stalks per mile) | | Field stands (stalks per acre) | |
|---|---|---|---|---|
| | Range | Average | Range | Average |
| Absent | 0 to 50 | 0 | 0 to 100 | 0 |
| Poor | 50 to 700 | 300 | 100 to 1,000 | 500 |
| Sparse | 700 to 3,000 | 1,500 | 1,000 to 5,000 | 2,500 |
| Medium | 3,000 to 8,000 | 5,000 | 5,000 to 10,000 | 7,500 |
| Good | Over 8,000 | 10,000 | Over 10,000 | 15,000 |

FIGURE 3. A roadside stand with 95 stalks in the strip about 2 yards wide and 1 rod long, equivalent to 30,000 stalks per mile, 1½ miles west of Fallowfield in Nepean Township, Carleton County. The larger plants are about 3 feet tall, producing from 8 to 22 pods each or an average of 12 pods per stalk. The soil here is heavy, the plants growing on the well-drained shoulder above a wet ditch. This patch would be rated as "good" density, however, the whole strip along this road was graded "medium" on account of some thin stretches.

FIGURE 4. A wild stand of milkweed on the heavy clay soil at Fallowfield Station near Ottawa. This field of about 6 acres in extent, has at times been cultivated, used as a meadow, and this year heavily grazed. Counts gave 19,250 stalks per acre, a high ("good") category of density. About 1 stalk in 20 is bearing pods on the average of about 6 per stalk. The roots are shallow, only about 2 inches deep on this soil, under a sod of timothy, couch grass and perennial weeds.

FIGURE 5. Milkweed in a pasture field at Ottawa along the Rideau River. This stand would be rated as "medium" density. It has been pastured for many years.

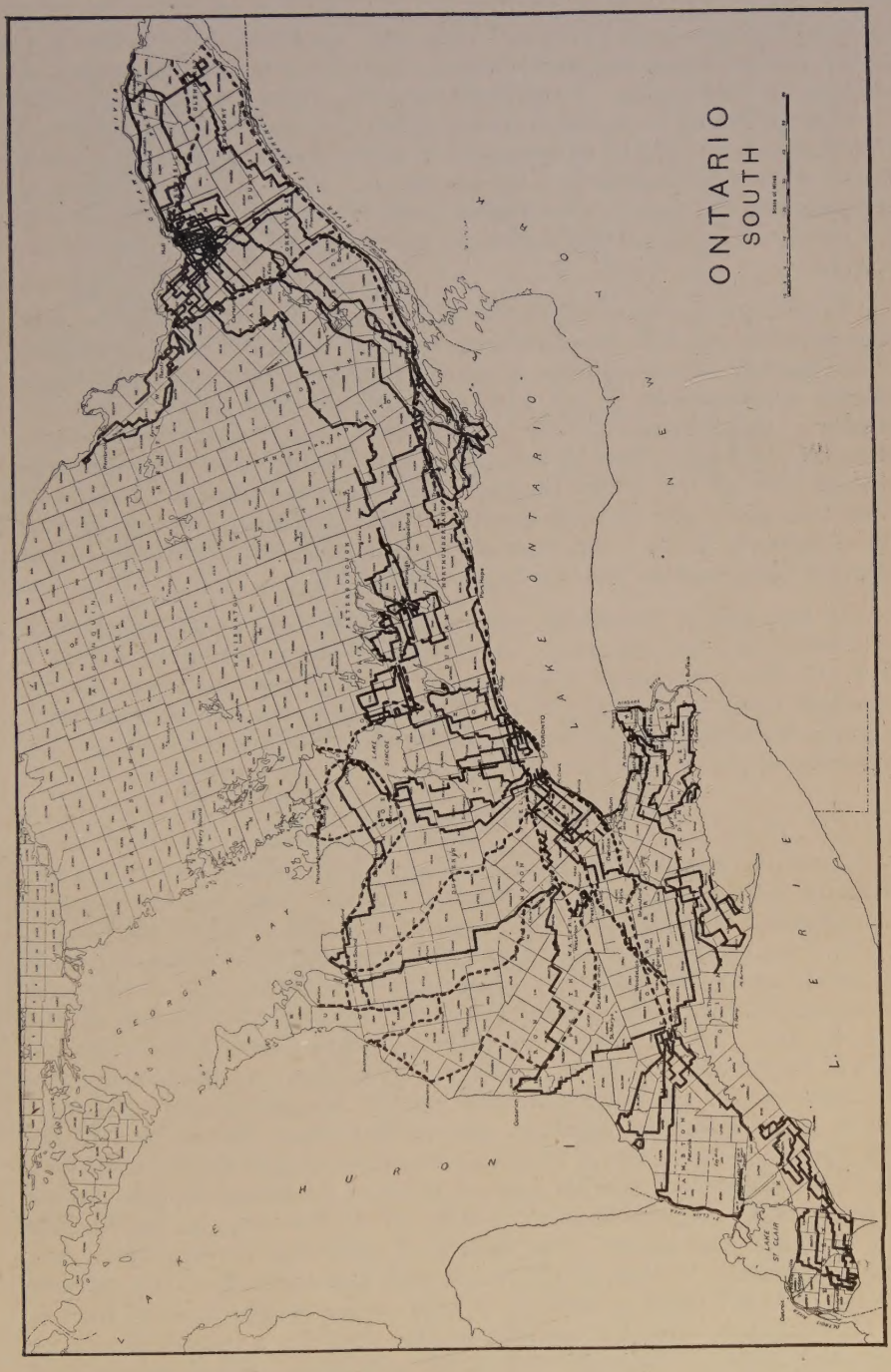


FIGURE 6. Survey routes in Ontario. Routes traversed by automobile are represented by solid lines; train and bus by broken lines.

Observations were recorded on maps carried along or in part in notebooks. Sheets of the National Topographic Series of the Department of National Defence, scale 1 mile to 1 inch, were used folded conveniently to receive entries delimiting each change of roadside density as passed by and locating field stands. From these roadside entries the various densities were later coloured in permanently according to a scale of colours adopted. They were then ready to be measured in miles of each by means of a calibrated wheel and summarized by townships. Field stands, indicated as to position on the map by a numbered circle, were at the same time entered in a notebook to show judged acreage, density grade and character of the habitat.

From these two sets of data the desired single figure for stalks per mile from roadside and field stands was obtained by addition. Then, dividing by the number of survey miles, a relative figure was obtained which made possible a comparison of one with another regardless of its size or the number of miles travelled. For the township or other unit adopted, these relative figures could then be arranged in descending order of density and grouped into the five grades shown in the legend of Figure 7. It is to be noted that the limits of these groups do not necessarily correspond with any of those in Table 1 but were derived more or less arbitrarily to put about an equal number of townships in each.

As mapped by townships, centres of high, diminishing outward to lower densities, are apparent. A generalized presentation in Figure 8 serves to blend townships missed entirely with those adjoining, and to focus still more the results obtained. Local extremes due to inadequate sampling are smoothed out in accordance with judgments based on personal knowledge of the situation or on replies to questionnaire enquiry.

## MILKWEED POPULATIONS

Coming to a scrutiny of the map figures it can be seen that in Ontario (including Quebec parts of the Ottawa Valley) there are three major concentrations of milkweed. That in eastern Ontario, roughly the Ottawa Valley, is segregated from the others rather sharply by the wooded Laurentide shield on the northeast, and the rugged Frontenac axis of the same Precambrian formation reaching south to the Thousand Islands of the St. Lawrence. To the west, from end to end of the Trent Canal zone and north of the narrow lake plain, concentration is high. In the remainder of the area surveyed the most significant feature uncovered appears to be a paucity of milkweed from almost the whole plateau above the Niagara escarpment line to the Bruce Peninsula and extending to the St. Clair flats and Erie shore, where another concentration of milkweed occurs. Below the escarpment, the level land north and west of Toronto, and the Niagara fruit belt, are also sparse. The dearth of milkweed in this region was especially notable in some of the heavy soils encountered. To a remarkable degree, also, there is conformity with earlier geological shore lines of the Great Lakes. Further analysis of this and other factors should be profitable, although perhaps, only after still more critical surveys. A fourth zone, of less extent, on either side of the city of Toronto is to be noted.

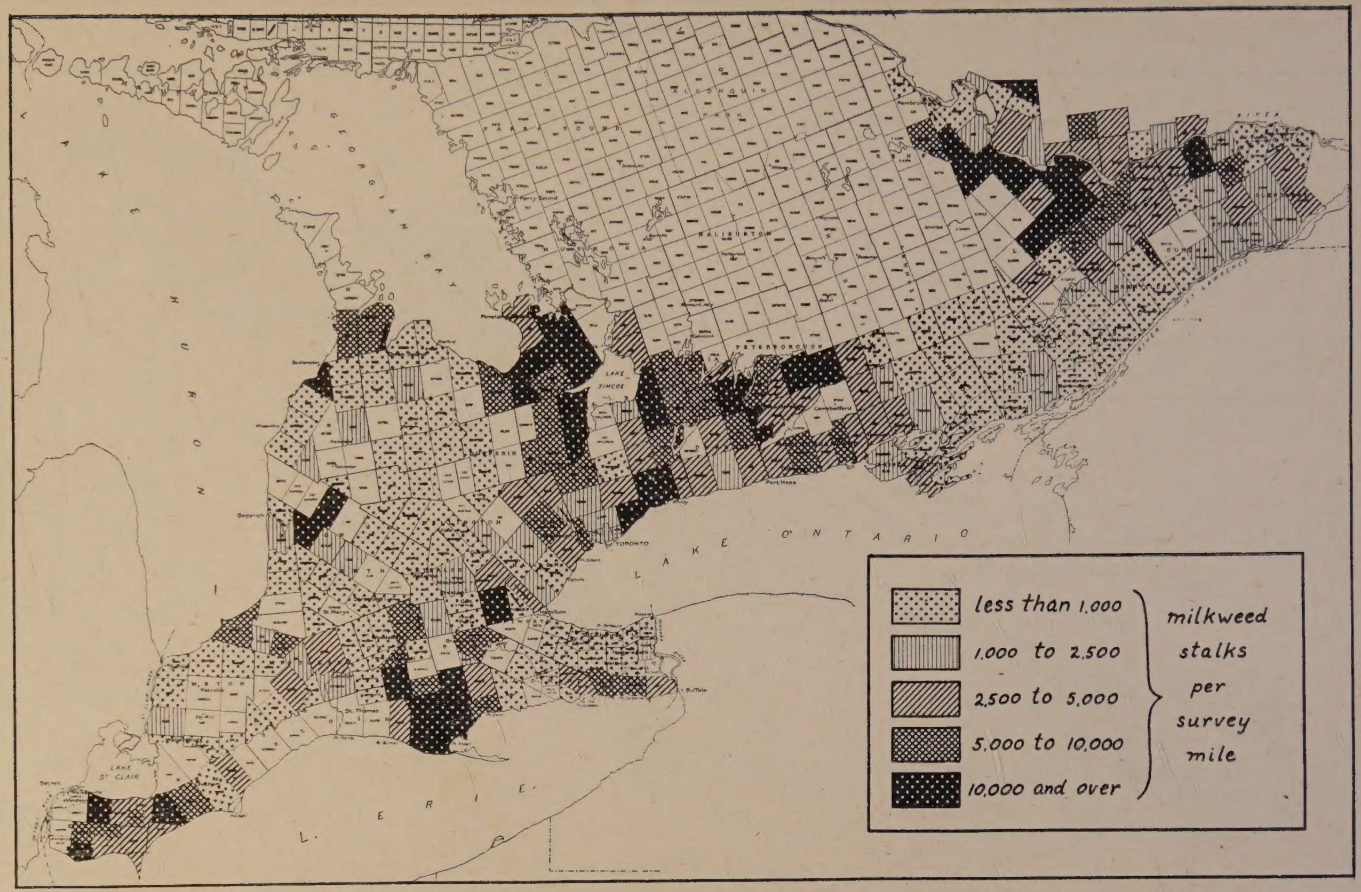


FIGURE 7. Milkweed density by townships based on averages of roadside and field stands as observed while travelling routes indicated in Figure 6. Unshaded townships were not traversed.

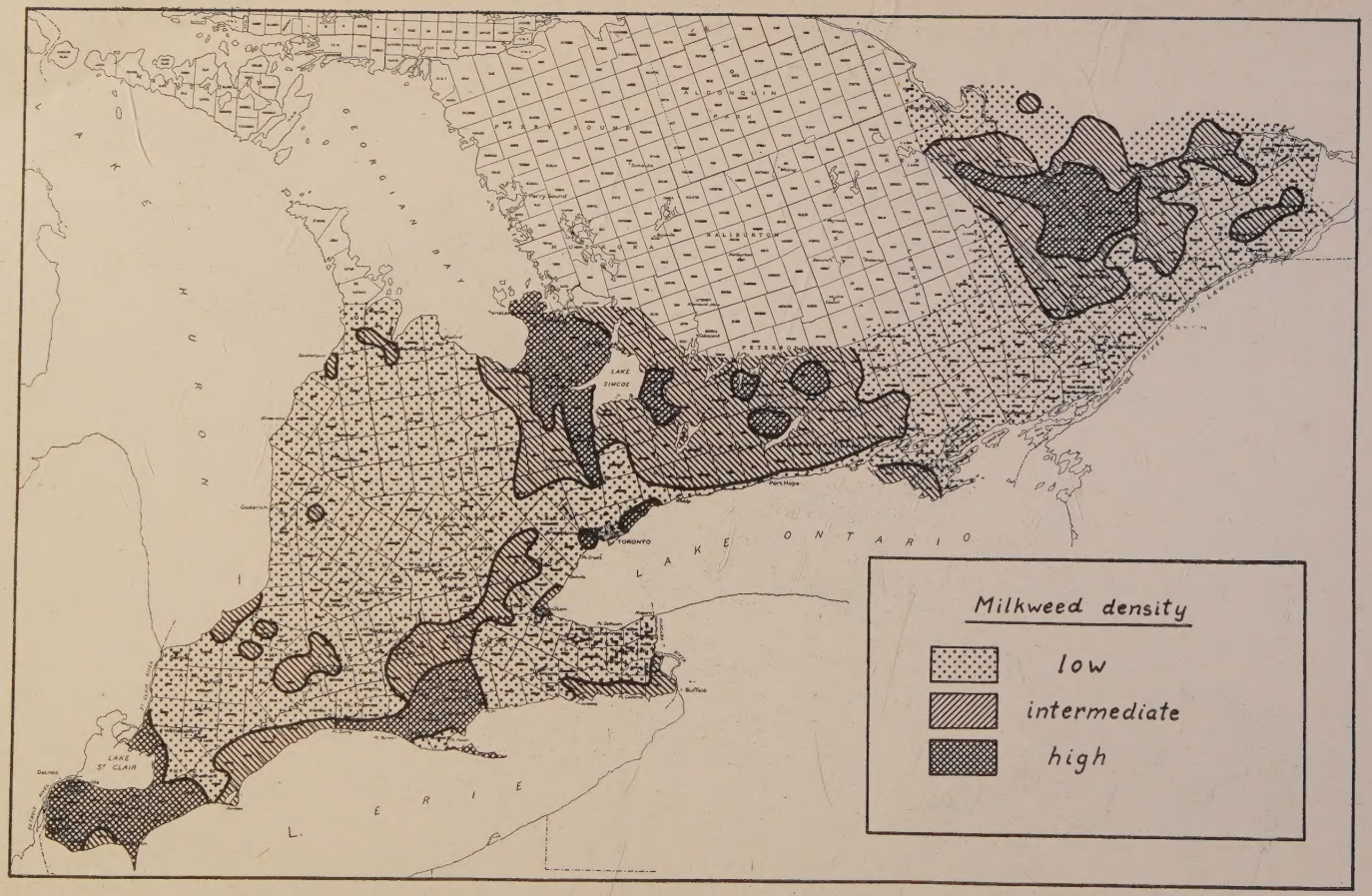


FIGURE 8. Generalized map of milkweed populations of various densities based on the township averages as indicated in Figure 7.

Some faults inherent in the sampling method may be pointed out which, while not affecting seriously the broad outlines, have a bearing on the more local applications. For instance, the border townships along the Detroit River, central Lambton County, and the greater part of Elgin's narrow strip were missed in the survey, and in the generalized map had to be merged with adjacent findings in the light of probability. Again, Morris and Hullett Townships in Huron County are an example of how a concentration of about 35 acres of milkweed around the town of Blyth, situated on the boundary between them, could create a "high" for two townships not in keeping with what was found for the region generally. A similar instance is afforded by Rockland in Russell County, and others in various places both East and West. When sampling is represented by an insufficient number of survey miles a single large field of milkweed can throw the stalk averages out of proportion to the general density over the area. In other cases, as in Canboro Township of Haldimand County, and Wilberforce Township, Renfrew County, the averages, based on observations along marginal strips, are not truly representative. Moreover, it is felt that the results in an area like North Simcoe might be interpreted more conservatively than they were in view of the fact that more than usual of these dense stands were of dwarfed and sterile plants. The survey recorded stalks regardless of vigour or productivity.

## DISCUSSION OF DISTRIBUTION

The rather striking segregation of milkweed populations in Ontario into three distinct regions, and these already foreshadowed in the preceding weed surveys (1), provides a starting point for consideration of factors which could be responsible. No simple hypothesis proves to be adequate throughout but several things are at least contributory.

### *Topography and Drainage*

While topography varies only within rather narrow limits the following physiographic features can be distinguished: (1) lowlands of the St. Lawrence and Ottawa rivers; (2) Frontenac spur and Laurentian shield north of Ottawa Valley; (3) plain between Frontenac spur and Niagara escarpment, including a rough glaciated region of deep morainic deposits through Peterborough to Simcoe County; (4) plain above Niagara escarpment in places exceeding 1,000 feet above sea level, but including flat lowlands to the southwest. Thus it is seen that the three milkweed belts are comprised within two of these tracts and the least elevated part of the third, while the greater part of the western Ontario peninsula and the Frontenac axis lie between them.

Altitude, as such, need not be stressed as a factor as the drainage basins concerned seem to be of more significance. Some of the lowlands which are sandy, as the lake shore deposits in Essex County where milkweed populations run from 10,000 to 20,000 stalks per mile, in western Norfolk and extending into Elgin County, in Welland County along the shore, on the Lake Huron shore, and the Lake Ontario shores at Hamilton Bay and in Prince Edward County, are all well enough drained and support milkweed. But more generally lowlands are level, clay lake sediments, and are poorly drained unless of loose alluvial deposits or when served by

drainage canals. Apart from such exceptions they are deficient in milkweed, as very commonly in northern Essex and Lambton Counties, north and east of Toronto, and in much of the lowlands west of Montreal.

Uplands are of irregular topography with irregular drainage. The drumlin region in Durham, Northumberland, Peterborough, and Victoria Counties, and the morainic deposits west of Lake Simcoe give a great variety of soil and drainage types with many of them favouring milkweed. On the other hand the equally irregular Frontenac spur and Laurentian uplands, with their numerous lakes, are not generally favourable to milkweed, but for other reasons than drainage. The elevated plateau of the western Ontario interior is a flat and heavy terrain almost devoid of milkweed. Topography, as influencing drainage, undoubtedly has significance.

*Climate and Drainage*

Precipitation in these parts of Ontario is mostly between 30 and 40 inches annually. Small areas in the Ottawa Valley, between Georgian Bay and Lake Ontario, and at the extreme southern tip of Ontario receive less than 30 inches annually. These, roughly, are areas of milkweed abundance. The correlation may be mere coincidence, but it is of interest to note that the plant has an eastern limit of abundance toward the more humid climate of the Maritime Provinces. To the west common milkweed extends into eastern Manitoba in the zone of 20 to 30-inch precipitation, mostly in stream valleys where soil moisture is assured, but only sparingly beyond at less than 20 inches. There showy milkweed (*Asclepias speciosa* Torr.) and dwarf milkweed (*Asclepias ovalifolia* Dec.) largely supplant it.

As altitude was not, probably, the significant feature in topography so precipitation within broad limits would seem to be less important than soil drainage. At each of the three points with slightly less precipitation, drainage happens to be generally satisfactory. On the clay plains of western Ontario, and even on Rubicon sand in Russell County in eastern Ontario, both poorly drained because of level topography, milkweed is almost absent. For a part of the season these clay soils are wet almost to the surface (when not artificially drained) and root growth is forced to shallow levels. Droughts following, opening up soil cracks, both injure roots and deprive them of water. The species is not averse to moisture but must have sufficient soil aeration. The thick horizontal root apparently cannot endure a water-logged condition. In deep sandy soils the same trouble can be encountered in abnormal seasons when, as in the very wet August of 1943 with 9 instead of the normal 3 inches of rain at Ottawa, plants with roots growing deep in the open substratum were found killed in September, presumably through the raising of the water table above the level at which they had established themselves. It is a general observation that saturated swamp soils do not support milkweed although the well-drained road grades crossing them may be covered profusely. Once only did there appear to be a serious exception to the rule: at Walkerton, in Bruce County, seepage from a dam, keeping lower ground permanently in a state of moisture to maintain typical swamp vegetation of alder, willow and the like, included with them a fair amount of milkweed. It was growing, perhaps in competition for light, to record heights of 6 and 7 feet and bearing pods normally. Investigation probably would have disclosed

shallow-rooting reaching into hummocky spots to escape the saturation just below. In this situation too, there would not be the violent changes of water level involved in the other instances cited; fluctuation, seasonally or from year to year, rather than a higher or lower uniform level (above saturation) appears to be the critical factor.

*Soils*

On the whole there is considerable support for the view that soil moisture and drainage are of consequence. Whether soil itself (its texture, structure, acidity, fertility, etc.) is important has not been easy to determine. Relationships have not been clearly worked out because soil maps were not everywhere available and the survey is still too general.

Excellent stands of milkweed are to be found on soils of any textural group—sand, clay or muck, particularly when they tend to a loamy character, and providing they are well drained. The plant seems responsive to fertility. When sands are leached of fertility the plants, although present, are not luxuriant. This was apparent on sandy ridges and plains in Simcoe County and on extensive tracts near Otter Lake, in Pontiac County, Quebec. On the other hand when fertilized, even with commercial fertilizer as in the sandy tobacco fields of Norfolk County, stands were strong, where otherwise not so. Clay soils are naturally more fertile but, as illustrated in northern Essex County and up the Ottawa River, are productive of milkweed only when well-drained and agriculturally fit. Some of the best milkweed seen, in the Ennismore and Ipperwash districts, was on land approaching the muck type, which had undergone drainage and amelioration to a state of high fertility. Confusing the issue, though, is the fact that frequently in the Ottawa district fields of well-podded milkweed can be seen on almost bare, irregularly eroded limestone strata, the stalks emerging from deep fissures of the rock about as well as from the pockets of soil forming in every depression. Just what is the provision for moisture here is not clear, unless efficient root storage carried the plant through intermittent drought.

Soil derivation and the influence of underlying rock is of interest. Limestone is frequently, in eastern Ontario, quite shallow under flourishing stands of milkweed, and underlies much of its territory also in central Ontario, but that at the lower extremity of the province grows on lake deposits of considerable depth over the underlying Devonian limestones and shales. Outcrops of crystalline limestone within the Precambrian formation are also commonly milkweed areas. Only for the exception of southern Ontario this might argue limestone or alkaline preference of the species, but tests made of acid tolerance varied between pH 6.6 and pH 5.7, or scarcely neutral. More acid soils such as would be derived from granitic rocks, or from sandstone or clay soils sour from poor drainage, are definitely uncongenial. Soils of limestone origin on the other hand, are not necessarily highly alkaline, since leaching in a climate such as this ordinarily reduces them to near neutrality or even to acidity. The milkweed concentrations near Ottawa and in central Ontario are both on predominantly neutral soils. Roadsides, otherwise acid, are doubtless often rendered congenial to milkweed by pulverization of the limestone material applied to roads.

Absence of milkweed from certain shallow soils over level limestone strata in Prince Edward County and adjoining Frontenac County, and around Ottawa, might be attributed to their lack of moisture-holding capacity between rains, rather than to any unfavourable pH due to proximity of the limestone.

*Light*

Only rarely has milkweed been found in shaded conditions. Over most of the surveyed area cleared and wooded portions are fairly uniformly interspersed in the proportion of 20% (or less in Old Ontario alone) of woodlot to 80% of cleared or grass and marsh land (Census, 1931), and milkweed was notable only for its scarcity through these wooded stretches. Through the Lennox-Frontenac hinterland this would share with the granitic forest floor in responsibility for the dearth of the plant. In diffused light it may struggle upward into lanky growth, but more dense shade dwarfs it and soon extinguishes life.

*Cultural Considerations*

Under this broad head may be reviewed most of the man-made conditions which undeniably enter into any appraisal of the situation. It is generally conceded that *Asclepias syriaca* is native to North America, although originally it would be less widely distributed than now. Forest cover and water-saturated soils in pre-settlement times were the restricting factors to a greater degree than now. Savannah openings in the forest, sandy river flats and shore dunes and the plains westward would be the probable sources populating its present range. Certain observations of the survey suggest that migration eastward is still in progress. In the Eastern Townships of Quebec a tendency to isolation in pure clones along roadsides, in orchards and hay fields, rather than in pasture and roadside stands of continuous and intermixed clones, as in Ontario, might be interpreted as indicating recent establishment. Farther east, while there are early records from Quebec city, Matapedia, Fredericton, etc., others of the comparatively few and localized infestations are probably more recent. In Nova Scotia the plant was unknown until 1930, since which date almost an axis of stations has been stretched through the Annapolis Valley, with a report from Cape Breton. There are still no records from Prince Edward Island and the mainland opposite.

Climatically, and with regard to the other factors discussed, all of settled eastern Canada should come within the bounds of milkweed tolerance. It appears merely a matter of time and the operation of requisite agencies of dispersal to bring much of the remaining area under invasion. It may be doubted, however, that either here, or again in Ontario, will invasion be such as was depicted in a paper read by Alexander Kirkwood before the old Ottawa Natural History Society in 1867 when he said: "The milkweed is well known to almost every farmer and next to Canada thistle, is viewed by him as his greatest pest."

Kirkwood's picture may be less overdrawn than might be thought. If it is remembered that the best of Ontario farm lands must have passed through the cruder phases of development which still mark areas of marginal utilization owing to their topographically broken and less arable character,

the situation will be clearer. Forgetting for the moment altitude, precipitation, soil, and even drainage and light conditions as separate factors, a glance at the maps will reveal considerable association of high milkweed density with agricultural conditions only fair to marginal. Much of eastern Ontario's "high" is over shallow rock outcrops as contrasted with the less broken St. Lawrence lowlands east of there. The Trent Canal system is much less amenable to high tillage than the broad stretches of farm land north and west of Toronto. Above the Niagara escarpment, again, is a tract of more than 14,000 square miles, mostly under highly developed agricultural use, with milkweed at its best only in the sandy parts of Norfolk and Essex Counties devoted to tobacco growing under heavy reinforcement of artificial fertilizers, or even withdrawn from cultivation for reforestation. The point is that flourishing land use, whatever else may be conducive to milkweed, holds it in check, or strikes a balance between tendencies at least. As land clearance admitted, so agriculture finally stabilized the weed at its present more or less innocuous state in this or that belt.

As a corollary to the foregoing the suggestion is offered that any prospective demand for milkweed products should look, not to lands already well enough utilized but to those less favourably placed as regards crop adaptation, accessibility to markets, or under other disabilities of the "back concessions." Having now more than their share of milkweed let them capitalize on it. Extensive areas between Peterborough and Georgian Bay, for instance, with dwarfed and largely sterile stands on land furnishing indifferent pasture would, there is reason to believe, be stimulated to heavier production of this plant (and grass) by means of sharp surface scarification of disc or cultivator in spring or after soaking rain. In some cases ploughing would be a warranted expense, judging by numerous instances where incipient stands from roots, almost suppressed in old sods, have been found restocking promptly those parts broken up for potatoes, buckwheat or other grain. Any programme of milkweed increase should consider well the merits of such root propagation before embarking on little understood seeding operations on better land.

With further reference to pastures it may be doubted that grazing has much direct controlling influence on milkweed. The trampling of live stock and consequent firming of the soil no doubt retards it somewhat. Sheep are reported to eat the young plants, and some "topping" was observed in a Gatineau County, Quebec, pasture carrying young cattle. Ordinarily, animals graze closely around, without touching, milkweed stems. Plants dried in hay, on the other hand, are reported by Michigan observers to be much relished. On the whole lands in permanent pasture, more than in any other use, were the sites of heavier infestation, in eastern and central Ontario particularly.

Before leaving the subject of man-made conditions attention should be drawn to a distinct concentration of milkweed commonly in the neighbourhood of towns and cities. Land previously well farmed, under the influence of rising property values, reverts to weeds and grass in subdivisions awaiting use as building sites; and meanwhile provides the zone of density which has enabled schools of suburban Toronto and other centres of population to share fully with more rural districts in the harvesting of

leaves or pods. This is clearly shown east and west of Toronto in Figure 8. The same effect northward is neutralized in the mapping by the inclusion of so much land still attractive for intensive agriculture. The contrast between the more settled shore strip through Scarboro and on into Ontario County, and that inland through Agincourt, is at least partially thus explained.

The cycle of change from cultivation, through weeds and thin grass, to a closer sward is illustrated also in the roadside surveys. Secondary and less used roads, where weeds and infrequent mowing leave debris to prevent formation of a close sod, were appreciably richer in milkweed than main travelled highways, whether these were seeded down or merely allowed to grass over with the help of closer attention. In several years' history of a meadow the same decline of the plant upon the formation of denser sward has been observed. Railway embankments, and rights of way cleared of rubbish by frequent burning were usually characterized by open sod frequented by milkweed.

Other forms of life, as insects and disease organisms, attack milkweed, but can hardly be credited with any great influence on either density or incidence. Virus disease, everywhere prevalent, results in a dense clumping of stems throughout the extent of the clone infected, together with yellowing or mottling, and finally deforming of leaves, stems and flowers, and little fruiting. Flagellates are reported as occurring generally in the latex of normal plants. In Renfrew County in a vigorous crop of milkweed, whole clones up to several square rods in extent were being defoliated prematurely by what was suggestive of bacterial disease. Aphids were often abundant rendering the plants sticky with their exudates. Two leaf-eating insects were common, and once a gregarious species of hairy caterpillar had nearly stripped the foliage. In one instance white grub injury to the root was found to be the stimulus to proliferation of shoots similar to that present with virus troubles. The milkweed caterpillar, larval stage of the familiar Monarch butterfly, is not the factor in defoliation that might be expected.

## ESTABLISHMENT AND DEVELOPMENT

An ordinary stalk of milkweed may produce several hundred seeds; and the stalks of an entire clone this number many times multiplied. The seeds with their floss are well equipped for wide distribution by wind so that every suitable site within its natural range should be well populated. This is far from being the case. In field work seedlings are rather infrequently found, and experience with seedings has not been uniformly successful. Without here going further into the bionomics of the seed it may be observed that its considerable size and lightness, even when detached from the floss, must operate to keep it from settling readily through surface material to a germination bed.

When a seedling has been successfully started a clone soon develops if conditions are good. Root extension starts in the latter part of the same year, even though new shoots do not reach the surface until the following spring. Once there is an ample foliage, surface root development becomes quite active, and plants by the second and third years from the seed, even if sown in rows, spring up indiscriminately all around and to some distance.

Numerous adventitious buds appear over all the root, the majority remaining dormant until space above or other conditions warrant their activity. The most vigourous stems arise from around the crown marking the previous year's growth. If these are lost by cutting, rub injury or disease, other buds, sometimes in excessive numbers, are called into activity. Flowering can occur in the first year but reaches its fulness only as reserves accumulate by about the third year.

The usual outcome of what has just been described is a colony of greater or less extent as soil conditions or inherent characteristics may have determined. Examination of a field shows that the stalks belong to relatively few clones, sometimes becoming intermingled, but recognizable by individuality of stature, leaf shape, colour of stalks and flowers, follicle type, etc. Each of these clones, unless root fragments were distributed by cultivation or other disturbance, had its origin from a single seed. Obviously the species is not greatly dependent for its persistence on continuous seed propagation, but has that alternative when unoccupied range is suitable and is not pre-empted by other vegetation.

## AN APPLICATION OF THE FINDINGS—POD YIELD

For use in the 1944 pod collection campaign the best available advance estimates of potential production were required, and such application of the survey figures as they would bear were made. County estimates were needed for this, therefore the township data for combined field and roadside stands were brought together for the larger unit. Only those counties were included in which sampling involved travelling 10% or more of the road mileage by automobile. Before including the field stands in the combined totals, however, they were weighted by three, which was considered a safe factor to extend observations from the road to the middle of any block of land and thus represent total milkweed population. The average per mile for these counties was then multiplied by the total mileage in roads, as supplied by the Ontario Department of Highways, to give the figures in the 4th column, Table 2. Further, a rough assumption of 1 pod for each 2 stalks, and 800 pods per bushel was made for the purpose of calculation. It is appreciated that in these particulars variation can be quite wide. As merely the best figures immediately available on yield, and for comparative rather than absolute use, they are set down as worked out in Table 2.

From Table 2 it would appear that Simcoe and Renfrew are the really high producing counties. Next in order are Peterborough, Ontario, York and Norfolk. All these are counties where field stands contributed heavily to the total, and the weighting to apply the field stand results to the whole area was additionally in their favour. Counties notably low are Lincoln, Haldimand and Halton in the west, and Lennox and Addington and Frontenac in the east.

While the actual figures of pod collection in 1944 are not available at the time of writing, some indication of the reliability of these estimates may be gained from the yields of dried leaves in 1943. The counties given a high estimate above, except Norfolk, provided over one-half ton of dried leaves, while the four named for low pod yield collected one-half ton or

less. Quite extraneous reasons for the relatively low leaf collection of only one-quarter ton in Norfolk are to be found. It is obvious that many things besides actual yield enter into the ultimate collection results.

TABLE 2.—MILKWEED DENSITY AND ESTIMATE OF POD YIELD FOR SOME COUNTIES IN ONTARIO

| County | Stalks per mile | | Miles of road in county | Total stalks (thousands)* | Green* pod yield (bushels) |
|---|---|---|---|---|---|
| | Roadside Stands | Field Stands | | | |
| Carleton | 1,665 | 5,850 | 1,298 | 24,900 | 15,600 |
| Essex | 3,000 | 3,760 | 1,176 | 16,800 | 10,500 |
| Frontenac | 360 | 370 | 1,121 | 1,650 | 1,030 |
| Haldimand | 720 | 870 | 757 | 1,850 | 1,150 |
| Halton | 700 | 575 | 573 | 1,900 | 1,180 |
| Hastings | 560 | 1,870 | 1,777 | 10,970 | 6,850 |
| Kent | 1,000 | 4,085 | 1,602 | 21,240 | 12,650 |
| Lanark | 1,600 | 5,200 | 1,216 | 20,900 | 13,000 |
| Leeds and Grenville | 760 | 450 | 1,660 | 7,300 | 4,570 |
| Lennox and Addington | 420 | 120 | 809 | 640 | 400 |
| Lincoln | 160 | 200 | 816 | 620 | 390 |
| Middlesex | 900 | 1,775 | 1,999 | 12,470 | 7,800 |
| Norfolk | 1,820 | 8,850 | 1,130 | 32,000 | 20,000 |
| Ontario | 1,161 | 12,760 | 1,381 | 54,400 | 34,000 |
| Peel | 500 | 3,625 | 724 | 8,240 | 5,150 |
| Peterborough | 1,240 | 17,300 | 1,122 | 59,700 | 37,300 |
| Prescott and Russell | 1,140 | 2,870 | 1,234 | 24,700 | 15,000 |
| Prince Edward | 370 | 2,000 | 547 | 3,500 | 2,200 |
| Renfrew | 800 | 17,640 | 1,914 | 102,800 | 64,260 |
| Simcoe | 1,470 | 16,340 | 2,369 | 119,600 | 75,750 |
| Victoria | 1,000 | 8,000 | 1,114 | 27,900 | 17,450 |
| Wentworth | 1,015 | 7,610 | 718 | 17,100 | 10,700 |
| York | 665 | 7,580 | 2,014 | 47,150 | 29,470 |

* Calculated as described in text.

## OTHER MILKWEEDS

Swamp milkweed (*Asclepias incarnata* L.) was in essentially the same Ontario range but occupying distinctly moister habitat. It was relatively uncommon in the eastern parts, more abundant in the Peterborough district, and reached high prevalence only in the southwestern peninsula. It was especially prominent in roadside ditches and other wet land in Kent, Essex and parts of Lambton Counties. Butterflyweed (*Asclepias tuberosa* L.) was found common only in sandy areas along Lake Erie and lower Lake Huron, rare and sporadic on sand plains elsewhere, and including one such in the Ottawa district. Sullivant's milkweed (*Asclepias Sullivantii* Engelm.) has been found at a few stations about Lake St. Clair, through the efforts in the past two years largely of Dr. W. Sherwood Fox. A few other species are known by herbarium specimens to be represented in southern Ontario counties. The pods of any which are not too rare may find their way into collections of common milkweed and without serious objection to such admixture.

## SUMMARY

A survey of density and distribution of the wild stands of common milkweed (*Asclepias syriaca*) in Old Ontario (including the Quebec side of the Ottawa Valley) involved 6,132 miles of travel by automobile and train. Roadside and field colonies were recorded by density grades and were then

combined to be expressed as stalks per mile of travel. Maps prepared by plotting these figures by townships provided a mosaic from which regions of high or low density are apparent. Survey technique has been detailed.

Three main concentrations of milkweed are: the Ottawa Valley mostly westward from Ottawa; the morainic belt extending through the Trent drainage system to Georgian Bay; and the southwestern peninsula bordering Lake Erie to Detroit River. Regions of light occurrence are: the Frontenac axis of Precambrian rock and borders; heavy soil plains of the ancient glacial lakes bordering the lower Great Lakes; and much of the extensive plateau between the Niagara escarpment and Lake Huron.

Factors most strongly influencing this pattern of distribution and density appear to be: soil drainage and aeration; absence of shading; and various cultural conditions. Topography has its bearing on drainage. Within wide limits precipitation is of minor importance except as, superimposed on soils, it helps to create a complexity of drainage types. On heavy lowlands plants are subjected to extremes of wetness and desiccation, while alluvium and sandy or loamy tracts remain more uniform. Soil texture otherwise is secondary to fertility and agricultural fitness. Neutral soil reaction supports milkweed better than an extreme of acidic from granitic or sandstone derivation; but limestone, at the other extreme, is commonly tolerable because leached out to leave but slight alkalinity.

Although perfectly at home in the best of farming land, milkweed finds its opportunity oftener in submarginal lands not so well utilized, or in suburban areas, on neglected waysides, or in fields reverting to sod or newly broken from sod. Otherwise, stiff sod, swampy and wooded areas chiefly restrict it. Being unpalatable to live stock it also escapes the grazing suffered by other weeds. While subject to attack by various parasites it is not seriously held in check by these.

Although a high production of plumed seeds provides for wide colonization, actual propagation is more efficiently accomplished by the fleshy creeping roots.

The survey, besides helping to clarify understanding of environmental relations, has provided some guidance for pod collection activities through advance estimates of potential yield from each district.

## ACKNOWLEDGMENTS

As one phase of milkweed investigations under the general direction of Dr. H. A. Senn, Associate Botanist in the Division, the project has had his constant encouragement and help. In field work Agricultural Representatives of the Ontario Department of Agriculture have frequently been of assistance, as also Mr. William Newman in the Peterborough area, President W. Sherwood Fox of the University of Western Ontario in that part of the province, Mr. W. S. Rowe, Guelph, in western Ontario, and Major L. I. Johnson, Ottawa, in eastern Ontario. To these, and others who have been helpful, sincere thanks are extended.

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# A NEW LETHAL IN SHEEP

## NERVOUS INCO-ORDINATION OR PARALYSIS AT BIRTH[1]

K. RASMUSSEN[2]

*Dominion Experimental Station, Lethbridge, Alberta*

[Received for publication December 29, 1944]

For a number of years sporadic cases of paralysis in new born lambs have been noticed in the sheep flock at the Dominion Experimental Station, Lethbridge, Alberta. No special significance was attached to them as they were considered to be individual abnormalities. However, in 1942 a concentration of these cases was observed in one of the four inbred lines of Canadian Corriedales in the flock, and no cases occurred in any of the other breeding groups. Consequently it appeared that there was a genetic basis for this abnormality as all the sheep were managed as one flock except during the breeding season when breeding groups were segregated. During the breeding season all sheep were handled and fed in a similar manner so it was reasonable to assume that the condition was not dependent on environmental or nutritional factors.

### REVIEW OF LITERATURE

The most recent review of lethal and sublethal characters in farm animals by Lerner (1) lists only two abnormalities in sheep, namely, muscle contracture and paralysis, that indicated a possible similarity to the condition observed in the Lethbridge flock. Roberts (2) originally reported on the condition of muscle contracture in new born lambs. The main characteristic of this abnormality was that the limbs were rigidly fixed, with only a small amount of movement at the joints. Zophoniasson (3) reported on the condition of paralysis in Icelandic sheep in which the affected lambs were lame at birth and with inco-ordination in the rear quarters. The description given in his report indicated that the condition was not similar to that reported in this paper.

### DESCRIPTION OF ABNORMALITY

Observation of cases occurring in 1942 first led to the belief that the condition was readily recognized but in subsequent years it has become evident that a considerable range exists in the severity of the affliction so that in some cases the condition may be overlooked. In what may be considered to be typically severe cases the lambs are unable to get to their feet without assistance and cannot stand even when placed on their feet. Another characteristic of the severe cases is that there appears to be a muscle contracture causing a concave curvature of the back when the animals are lying down and a tendency for the neck to be flexed so that the head is drawn sideways from the normal plane. Furthermore, there is usually a more or less continuous muscular movement causing a twitching and kicking of the legs.

[1] Contribution from the Experimental Farms Service, Dominion Department of Agriculture, Ottawa, Canada.

[2] Head, Animal Husbandry Section, Dominion Experimental Station, Lethbridge, Alta.

From the severe symptoms there is a gradual diminution to the stage where the affected lambs cannot be distinguished with accuracy from lambs that may be weak at birth for other reasons. In some mild cases the lambs appear to be normal but as they develop and begin to move about it is noticed that their gait and stance are abnormal. The most noticeable feature of such cases is that the legs, particularly the hind legs, are not properly drawn under the body and this gives the lambs a spraddled appearance. Lambs affected in this manner have difficulty in walking as the legs are dragged rather than lifted in the normal manner.

The severely affected animals usually die within 48 hours of birth, mainly because of starvation brought about by their inability to stand and suckle. In 1942 one severely affected lamb was assisted for a couple of days and then improved to the point where it could get up and move around with some difficulty. For about a week it appeared to be making progress but then it began to weaken and finally died on the fourteenth day. Figures 1 and 2 (Plate 1) show this lamb in characteristic poses two days before death. It could drag itself along by using the front legs but had no power in the hind quarters.

Figures 3 and 4 show a lamb that was quite severely affected but was able to move about though it later died. These illustrations show the extreme condition of the abnormal stance of lambs affected but able to walk.

A lamb with slight symptoms is shown in Figures 5 and 6. This lamb was not observed to be abnormal until it was about a month old but then it was seen to always fall behind when the flock was being moved. Further observation showed that it had the abnormal gait that since has been found to be characteristic of the milder cases. The pictures do not show extreme abnormality but do indicate an abnormal position of the legs when the lamb is standing.

## BREEDING HISTORY

Since this condition first was noted in 1942 a study has been made of the possible hereditary nature of the condition and while this study is not completed it was thought that the material might be of sufficient interest to justify its publication.

The inbred line in which the abnormality first was observed is designated as Line 4. It was set up in the fall of 1934 along with other lines in the Canadian Corriedale flock developed at this Station. All lines had the same origin and had been flock mated up to that time. The division of ewes into lines was largely on a random basis and the original sires were selected from males available in the flock. The lines have been kept intact since their inception with the exception that one line was discontinued and the ewes divided among the remaining lines. Furthermore, one of the original lines has since become so large that it has been divided into two sub-lines.

## CAPTIONS

FIGURES 1 and 2 show a lamb so severely paralysed that it was unable to stand. Position of hind legs shows complete lack of control. Figures 3 and 4 show the abnormal stance of a lamb severely paralysed but able to walk. Figures 5 and 6 are of a lamb only slightly paralysed. Casual observation in a flock might not identify such a lamb.

The 1942 lambing record of Line 4, in which the abnormality appeared, is summarized in Table 1.

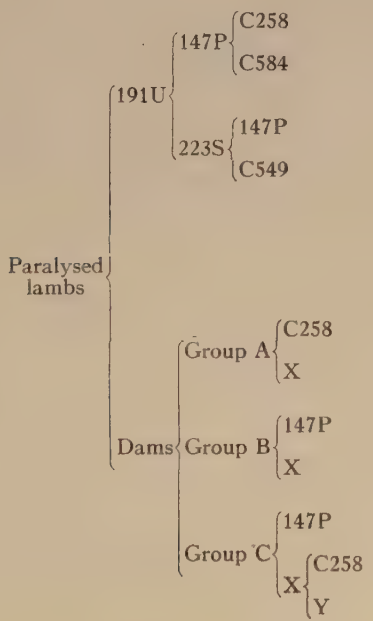
TABLE 1.—PROGENY RECORD FOR LINE 4 IN 1942

| Classification of lambs | Inbred | Non-inbred |
|---|---|---|
| Normal | 28 | 9 |
| Paralysed | 9 | 1 |
| Dead at birth | 4 | 0 |
| Died the first day | 0 | 1 |
| Killed at birth | 2 | 1 |
| Total | 43 | 12 |

The only data to which any significance can be attached are those classed as "normal" and "paralysed." Lambs dead at birth may or may not have been paralysed but this could not be determined. Those killed at birth were destroyed for other reasons having no apparent bearing on the present case.

The ratio of 9 paralysed to 28 normal in the inbred group definitely points to a genetic factor of a recessive nature. The single case among the non-inbreds need cause no particular disturbance as the gene responsible may well have been in the original stock from which the line was segregated.

The relationship of the various inbred paralysed progeny is shown in the accompanying chart.



This chart shows that all the paralysed lambs were sired by the same sire (191U) and the dams of the paralysed lambs were closely related to this sire.

## EXPERIMENTAL MATINGS

In order to study further the inheritance of the abnormality a special breeding group was set up in the fall of 1942. In this group all ewes that had produced paralysed lambs in Line 4 were mated back to the sire, 191U, that had produced paralysed lambs in this line. These same matings were continued in 1943 with the remaining ewes. A summary of the lambing results for these matings in 1942, 1943, and 1944 is given in Table 2.

TABLE 2.—PROGENY RECORD OF EWES THAT PRODUCED PARALYSED LAMBS WHEN MATED TO RAM 191U

| Ewe | 1942 | | 1943 | | 1944 | | Total 1942, 1943, 1944 | Total 1943, 1944 |
|---|---|---|---|---|---|---|---|---|
| | Sex of lamb | Class* | Sex of lamb | Class | Sex of lamb | Class | | |
| 156S | M | P | M | P | F | N | — | — |
| | — | — | M | N | — | — | — | — |
| 185S | M | P | F | N | M | N | — | — |
| | — | — | M | N | M | N | — | — |
| 223S | M | N | M | P | M | N | — | — |
| | M | P | M | P | M | N | — | — |
| 41T | M | P | M | P | M | N | — | — |
| 95T | P | P | M | P | M | P | — | — |
| | — | — | ? | P | — | — | — | — |
| 103T | M | P | — | — | F | N | — | — |
| | M | P | — | — | F | P | — | — |
| 212S | F | P | — | — | — | — | — | — |
| Total lambs | 9 | | 9 | | 9 | | 27 | 18 |
| Paralysed | | 8 | | 6 | | 2 | 16 | 8 |
| Normal | | 1 | | 3 | | 7 | 11 | 10 |

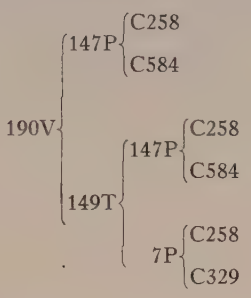
* P = paralysed.
N = normal.

Twenty-seven lambs were produced by these ewes during the three years but in view of the fact that the ewes originally were selected because they had produced defective lambs the 1942 data cannot be used in the present analysis. In 1943 and 1944, eighteen lambs were produced of which 8 were classed as defective and 10 as normal. It should be emphasized, as indicated earlier, that some mild cases of the defect may have escaped attention so the classification may not be completely accurate.

The data in Table 1 indicate that the defect is conditioned by a recessive gene and this is supported by the data in Table 2. However, the results from the special breeding group indicate that this may not be a simple case of a homozygous recessive condition. Undoubtedly there are

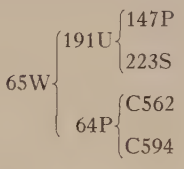
modifying genes involved and there is some evidence to show that possibly the milder form of the defect exists in individuals that are heterozygous for the causative gene. A definite decision on this cannot be made on the basis of available data.

Some supplementary evidence is available from the matings in Line 4. The ewes that did not produce paralysed progeny in this line in 1942 were continued in the line and mated that fall to ram 190V whose pedigree is as follows:




This is the same pedigree as that of 191U with the exception of the maternal section of the dam. In the 1943 progeny from this sire no paralysed lambs occurred. This would indicate either that this sire was free from the gene for the defect, or that all the heterozygous ewes had been removed from the group. This latter assumption is logically unsound and furthermore has been proven incorrect by later matings.

In 1943 a third ram, 65W, was used in Line 4. His pedigree is




On the maternal side he had no immediate relationship to the other rams, but apparently he had received the gene from his sire as he produced three paralysed progeny in the 1944 lamb crop resulting from this mating.

## DISCUSSION

While the breeding experiments are incomplete and not fully critical, the data support the assumption that the condition of paralysis observed in new born lambs has a hereditary basis and is not caused by specific environmental or nutritional factors. The simplest explanation of its mode of inheritance is that it is conditioned by a recessive gene with modifiers that determine the degree of expression. It might be assumed that the more severe cases are homozygous recessive animals whereas the heterozygotes would exhibit the less severe manifestations of the condition. The observations made on new born lambs have not been sufficiently critical to provide adequate data on which to base definite conclusions in this regard. Furthermore, breeding tests would have to be conducted to verify such a hypothesis, and this has not been possible up to the present time.

## PRACTICAL SIGNIFICANCE

Any abnormality that results in death of lambs is of definite practical importance. Consequently the paralytic condition described in this paper must be given serious attention wherever it may appear. It is by no means self eliminating even though all the homozygotes may die and a number of heterozygotes be eliminated through culling. It may be possible eventually to learn to detect all affected animals while they are young and thus eliminate them from the flock, but further study will be required along these lines. Until this can be determined the incidence of the characteristic can be kept at a minimum by systematic culling of all abnormal animals and by disposing of all breeding stock, that through a breeding test, have been shown to be carriers of the gene.

## SUMMARY

A lethal deformity of new born lambs is described. The main characteristic of this abnormality is inco-ordination of muscular action causing the lambs to be unable to stand. Muscle contracture in the muscles of the back and neck was observed in some cases. Also in some cases a continuous involuntary muscular activity caused the legs to constantly twitch and kick.

The characteristic appeared sporadically before inbreeding was started in the flock and then it concentrated in Line 4. The evidence from the breeding history indicated that the deformity was conditioned by a recessive gene.

Further evidence from experimental matings substantiated this but also showed that apparently modifying genes were at work as various degrees of the abnormality became noticeable.

In typically severe cases, assumed to be homozygous, death ensued within 48 hours because of starvation as the lambs were unable to stand. Milder cases survived but were characterized by abnormal gait and stance.

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# STUDIES ON THE OPTIMUM NUTRITION OF FLUE-CURED TOBACCO[1]

E. T. McEvoy[2]

*Experimental Farms Service, Ottawa, Ontario*

[Received for publication January 25, 1945]

While the nutrition of the tobacco plant has been studied extensively in sand and water culture, very little work has been done concerning the concentrations and relative proportions of all six major nutrient elements most favourable for the development of the plant. Therefore, a series of experiments was conducted at Ottawa to determine the optimum major-element requirements of flue-cured tobacco in sand culture and the effect of the nutrient ions on the growth of the plant.

A multiple-salt system of nutrient solutions was used. The solutions were devised from the triangle of Shive (2), using the system presented by Beckenback *et al.* (1). By this method, the relative concentrations of the cations and the anions are varied separately, providing a means of studying the physiological significance of the various ions. The cations are varied in multiples of a selected increment of the total concentration due to the cations, and the anions are varied similarly, the total number of increments for cations and anions per treatment remaining the same for each series of solutions. All nutrient solutions in each series are made up to approximately the same osmotic concentration. Likewise, the pH value varies very little throughout the series. Thus, these two factors, which are important in their influence upon the growth of the plant, are held constant, or nearly so, throughout the series.

A number of ionic proportions were selected and each of the cation proportions was combined with each of the anion proportions, making a series of nutrient solutions. These were arranged into a number of sub-series, in each of which the ratio of the cations was held constant but that of the anions was varied through the full range of proportions employed. A rearrangement of the same treatments gave additional sub-series, in each of which, conversely, the ratio of the anions was held constant but the cations were varied through the full range of proportions.

All solutions throughout the experiments contained the minor elements boron, manganese, and zinc at a concentration of 0.5 p.p.m. and iron at a concentration of 3.0 p.p.m.

Tobacco plants of the flue-cured variety White Mammoth were used in this experiment. The seedlings were grown for 8 weeks in sand culture in 3-inch pots, and all were supplied with the same nutrient solution. Shive and Robbins' formula II (3) was used for the first series, but was replaced in subsequent series by solutions developed in this investigation which produced better growth. The seedlings were transplanted to 3-gallon glazed jars, holding 40 pounds of washed, ground Nepean sandstone. One

[1] Contribution from the Tobacco Division, Experimental Farms Service, Dominion Department of Agriculture, Ottawa, Ontario.

[2] Agricultural Scientist.

plant was grown in each jar and the nutrient solution was supplied by the constant flow method from glass jars and capillary tubing, 1 litre per day being supplied to each plant.

The plants were harvested when mature, 2 weeks after topping. The stalks were cut off at the crown, the total fresh weights were determined and the stalks and leaves were separated for the determination of green weights and dry weights. The fractions were thoroughly mixed and aliquots were weighted for the various determinations. The samples were dried at a temperature of 70 to 80° C.

An analysis of variance, based on the total fresh weight data of the plants, was performed for each series. While a summary of this work is not presented, the difference between any two fresh weight means necessary for significance at the 5% level is appended to each table.

## RESULTS

*Series I*

In the first series, the plants were grown in 16 different nutrient solutions used by Beckenbach *et al* (1), each solution being made up to a total osmotic concentration of 0.5 atmosphere. The following chemically pure salts were used to balance the ionic ratios: $KH_2PO_4$, $CaH_4(PO_4)_2$, $MgH_4(PO_4)_2$, $KNO_3$, $Ca(NO_3)_2$, $Mg(NO_3)_2$, $K_2SO_4$, $CaSO_4$, and $MgSO_4$. The 16 treatments were divided into 8 sub-series. Sub-series 1 to 4 are characterized by fixed cation proportions and sub-series 5 to 8 are characterized by fixed anion proportions, each having variations in the relative proportions of the ions of opposite charge.

The seedlings were planted on October 21, 1939, and the plants were harvested on January 2, 1940. Three replications of each treatment were used.

The ionic proportions used and a summary of the results are presented in Table 1. The arrangement of treatments in sub-series 1 to 4 provides for a study of the influence on growth of the different anion proportions for each of the four fixed cation proportions. Considering the fresh weight yield in grams as the criterion of growth, greater growth was produced by treatments high in $NO_3$ and low in $PO_4$ and $SO_4$ than by any other treatment for each of the four fixed cation proportions. The 3-4-3 anion ratio ranked second in each case; however, in sub-series 4, it was not significantly different from the 8-1-1 anion ratio. The differences between the high $SO_4$ and the high $PO_4$ treatments were not statistically significant, except in sub-series 4 in which high $PO_4$ produced the greater yield. Over the range of anion variations employed, the greatest variation in yield was manifested when Ca was high, next greatest when the cation distribution was 3-4-3, and least when Mg was high.

In sub-series 5 to 8 of Table 1, the arrangement of treatments provides for a similar study of the data on the cation ratios. While the fresh weight yield associated with the cation ratio 3-4-3 ranked first in all cases, the increase in yield shown by this ratio over each of treatments 6 to 10 in sub-series 5, 16 in sub-series 6, and 5 in sub-series 7 was not statistically significant. When $NO_3$ was high, high yield was associated with intermediate Ca and high Ca in that order; when $PO_4$ was high, high yield was

TABLE 1.—YIELD DATA FOR SERIES I, ARRANGED IN SUB-SERIES 1–4 FOR COMPARISON OF ANION EFFECT UPON GROWTH AND IN SUB-SERIES 5–8 FOR COMPARISON OF CATION EFFECT UPON GROWTH

| Sub-series No. | Treatment No. | Cation proportions | | | Anion proportions | | | Mean fresh weight per plant | Mean dry weight per plant | Per cent dry weight | | | Water per gram dry weight | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Ca | Mg | K | $NO_3$ | $SO_4$ | $PO_4$ | | | Per plant | Stalks | Leaves | Stalks | Leaves |
| | | | | | | | | gm. | gm. | % | % | % | gm. | gm. |
| 1 | 1 | 3 | 4 | 3 | 3 | 4 | 3 | 128 ± 4.2* | 10.2 | 7.9 | 7.2 | 8.3 | 12.9 | 11.1 |
| | 2 | 3 | 4 | 3 | 1 | 1 | 8 | 94 ± 5.2 | 10.4 | 11.0 | 10.7 | 11.2 | 8.4 | 8.0 |
| | 3 | 3 | 4 | 3 | 8 | 1 | 1 | 181 ±23.3 | 16.9 | 9.4 | 10.0 | 9.1 | 9.0 | 10.0 |
| | 4 | 3 | 4 | 3 | 1 | 8 | 1 | 95 ±10.7 | 8.6 | 9.1 | 8.8 | 9.2 | 9.0 | 9.9 |
| 2 | 5 | 8 | 1 | 1 | 8 | 1 | 1 | 179 ± 2.0 | 20.7 | 11.5 | 11.0 | 11.8 | 8.1 | 8.5 |
| | 6 | 8 | 1 | 1 | 3 | 4 | 3 | 118 ± 7.0 | 12.7 | 10.7 | 11.2 | 10.5 | 7.9 | 8.5 |
| | 7 | 8 | 1 | 1 | 1 | 8 | 1 | 72 ± 3.2 | 8.4 | 11.7 | 11.8 | 11.6 | 7.5 | 7.6 |
| | 8 | 8 | 1 | 1 | 1 | 1 | 8 | 71 ± 3.0 | 8.2 | 11.7 | 11.2 | 11.9 | 7.9 | 7.4 |
| 3 | 9 | 1 | 1 | 8 | 1 | 1 | 8 | 74 ± 3.7 | 7.2 | 9.6 | 11.8 | 10.3 | 11.1 | 8.7 |
| | 10 | 1 | 1 | 8 | 3 | 4 | 3 | 122 ± 7.7 | 12.2 | 9.9 | 12.0 | 9.0 | 7.3 | 10.1 |
| | 11 | 1 | 1 | 8 | 8 | 1 | 1 | 143 ± 8.3 | 12.6 | 8.9 | 10.7 | 8.2 | 8.3 | 11.2 |
| | 12 | 1 | 1 | 8 | 1 | 8 | 1 | 77 ±14.5 | 8.6 | 11.2 | 11.7 | 10.9 | 7.5 | 8.2 |
| 4 | 13 | 1 | 8 | 1 | 1 | 8 | 1 | 69 ±10.5 | 7.6 | 11.1 | 10.4 | 9.8 | 6.1 | 9.2 |
| | 14 | 1 | 8 | 1 | 3 | 4 | 3 | 101 ±16.0 | 12.5 | 12.4 | 14.0 | 9.4 | 6.1 | 9.7 |
| | 15 | 1 | 8 | 1 | 8 | 1 | 1 | 109 ±15.2 | 12.5 | 11.5 | 12.8 | 11.0 | 6.8 | 8.1 |
| | 16 | 1 | 8 | 1 | 1 | 1 | 8 | 86 ± 5.3 | 8.1 | 9.1 | 11.8 | 8.2 | 7.5 | 11.2 |
| 5 | 1 | 3 | 4 | 3 | 3 | 4 | 3 | 128 ± 4.2 | 10.2 | 7.9 | 7.2 | 8.3 | 12.9 | 11.1 |
| | 6 | 8 | 1 | 1 | 3 | 4 | 3 | 118 ± 7.0 | 12.7 | 10.7 | 11.2 | 10.5 | 7.9 | 8.5 |
| | 10 | 1 | 1 | 8 | 3 | 4 | 3 | 122 ± 7.7 | 12.2 | 9.9 | 12.0 | 9.0 | 7.3 | 10.1 |
| | 14 | 1 | 8 | 1 | 3 | 4 | 3 | 101 ±16.0 | 12.5 | 12.4 | 14.0 | 9.4 | 6.1 | 9.7 |
| 6 | 2 | 3 | 4 | 3 | 1 | 1 | 8 | 94 ± 5.2 | 10.4 | 11.0 | 10.7 | 11.2 | 8.4 | 8.0 |
| | 8 | 8 | 1 | 1 | 1 | 1 | 8 | 71 ± 3.0 | 8.2 | 11.7 | 11.2 | 11.9 | 7.9 | 7.4 |
| | 9 | 1 | 1 | 8 | 1 | 1 | 8 | 74 ± 3.7 | 7.2 | 9.6 | 11.8 | 10.3 | 11.1 | 8.7 |
| | 16 | 1 | 8 | 1 | 1 | 1 | 8 | 86 ± 5.3 | 8.1 | 9.1 | 11.8 | 8.2 | 7.5 | 11.2 |
| 7 | 3 | 3 | 4 | 3 | 8 | 1 | 1 | 181 ±23.3 | 16.9 | 9.4 | 10.0 | 9.1 | 9.0 | 10.0 |
| | 5 | 8 | 1 | 1 | 8 | 1 | 1 | 179 ± 2.0 | 20.7 | 11.5 | 11.0 | 11.8 | 8.1 | 8.5 |
| | 11 | 1 | 1 | 8 | 8 | 1 | 1 | 143 ± 8.3 | 12.6 | 8.9 | 10.7 | 8.2 | 8.3 | 11.2 |
| | 15 | 1 | 8 | 1 | 8 | 1 | 1 | 109 ±15.2 | 12.5 | 11.5 | 12.8 | 11.0 | 6.8 | 8.1 |
| 8 | 4 | 3 | 4 | 3 | 1 | 8 | 1 | 95 ±10.7 | 8.6 | 9.1 | 8.8 | 9.2 | 9.0 | 9.9 |
| | 7 | 8 | 1 | 1 | 1 | 8 | 1 | 72 ± 3.2 | 8.4 | 11.7 | 11.8 | 11.6 | 7.5 | 7.6 |
| | 12 | 1 | 1 | 8 | 1 | 8 | 1 | 77 ±14.5 | 8.6 | 11.2 | 11.7 | 10.9 | 7.5 | 8.2 |
| | 13 | 1 | 8 | 1 | 1 | 8 | 1 | 69 ±10.5 | 7.6 | 11.1 | 10.4 | 9.8 | 6.1 | 9.2 |

* Difference between means necessary for significance at the 5% level = 11.8.

associated with intermediate Mg and high Mg; and when $SO_4$ was high, high yield was associated with intermediate K and high K. However, in the last case the difference did not reach the level of significance. The 1-8-1 cation ratio, in which Mg was at its maximum concentration, gave the lowest yield in each case, except sub-series 6, and resulted in necrotic areas on the leaves, the severity of the disorder being reduced considerably when the anion distribution was high in $PO_4$ and low in $NO_3$ and $SO_4$. It is clearly indicated that higher concentrations of Ca and K than of Mg are required to produce good growth. Variations in the cation proportions

resulted in greatest variation in yield when the anion ratio was high in $NO_3$ and in least variation in yield when the $NO_3$ concentration was low and that of either $SO_4$ or $PO_4$ was high.

The dry weight data show that high nitrate in the nutrient solution resulted in a low percentage of dry weight in the leaf. In sub-series 1, 2, and 3, the number of grams of water per gram dry matter in the leaf tissue was greater when the solution was high in $NO_3$ than when either $PO_4$ or $SO_4$ was high. Only in sub-series 4 in which toxicity of Mg was manifested were the data at variance with the general results.

Solutions with a high K concentration produced plants which were consistently higher in water content of the leaf fraction than did solutions with a high Ca concentration. In contrast, solutions high in Ca all produced leaf tissue with a higher proportion of dry matter than did any other solution. Comparisons in the stalk fractions, however, were not consistent in this respect. Also, the data from high Mg were quite irregular and did not reveal any definite influence of this ion on the relative proportions of water and dry matter in the tissue.

While only a small number of the possible solutions were tested in this series, sufficient information was obtained on the importance of the relative ionic concentrations to provide a bases for an extension of the work.

*Series II*

Using the same 9-salt system as in series I, 16 nutrient solutions were devised. Each ion was used at three concentrations, corresponding to multiples of the one-fourteenth increment of the total concentration. The amounts of Ca and $NO_3$ were varied within narrow limits around the concentrations of these ions which produced best growth in series I. Mg and $SO_4$ were used at comparatively low concentrations since growth was depressed in series I when Mg was high and, in less degree, when the amount of $SO_4$ was high as compared with $PO_4$ (sub-series 4). K and $PO_4$ were used at intermediate levels. The ionic ratios used, as given in Table 2, resulted in an increase in the total supply of ions in each solution as compared to that in series I; therefore, the osmotic concentration of each solution was increased to 0.7 atmosphere.

The seedlings were planted on March 1, 1940, and the plants were harvested on May 21, 1940, three replications of each treatment being used.

The yield data for this series are summarized in Table 2. The arrangement of treatments in sub-series 1 to 4 allows for a study of the comparative effect of the anion proportions and in sub-series 5 to 8 for a similar study of the cations. It is evident that the 9-1-4 anion ratio gave a yield which was significantly higher than that of any other treatment in sub-series 1 and 3, but was not significantly different from those of the 8-2-4 ratio in sub-series 2 and the 8-2-4 and 8-4-2 ratios in sub-series 4. The 6-3-5 ratio ranked last in each case, the difference between high and low nitrate being highly significant at all four cation proportions. The 8-2-4 anion ratio ranked higher than the 8-4-2 ratio in sub-series 1 and 2, while no significance can be assigned to the differences between these two treatments in the other cases. Such comparisons show that high $NO_3$ is associated with

high green weight; but, when used as 9 parts of the total anion concentration, the supply of this ion is sometimes in excess of that required for maximum growth (sub-series 2 and 4). However, this level of $NO_3$ is not above the optimum concentration as it did not reduce yield significantly in any instance. $PO_4$ is required at a higher level than is $SO_4$, a supply of 1 or 2 relative parts of the latter being adequate for good growth. It is not evident whether the decrease in yield when the amount of $SO_4$ was increased from 2 to 4 relative parts (sub-series 1 and 2), was caused by

TABLE 2.—YIELD DATA FOR SERIES II, ARRANGED IN SUB-SERIES 1–4 FOR COMPARISON OF ANION EFFECT UPON GROWTH AND IN SUB-SERIES 5–8 FOR COMPARISON OF CATION EFFECT UPON GROWTH

| Sub-series No. | Treatment No. | Cation proportions | | | Anion proportions | | | Mean fresh weight per plant | Mean dry weight per plant | Per cent dry weight | | | Water per gram dry weight | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Ca | Mg | K | $NO_3$ | $SO_4$ | $PO_4$ | | | Per plant | Stalks | Leaves | Stalks | Leaves |
| | | | | | | | | gm. | gm. | % | % | % | gm. | gm. |
| 1 | 1 | 6 | 3 | 5 | 6 | 3 | 5 | 765 ±32.5* | 109.1 | 14.4 | 14.1 | 14.6 | 6.1 | 5.9 |
| | 2 | 6 | 3 | 5 | 8 | 4 | 2 | 772 ±17.8 | 102.0 | 13.2 | 14.6 | 12.0 | 5.9 | 7.3 |
| | 3 | 6 | 3 | 5 | 8 | 2 | 4 | 862 ±31.6 | 103.8 | 12.0 | 14.1 | 11.1 | 6.1 | 8.0 |
| | 4 | 6 | 3 | 5 | 9 | 1 | 4 | 916 ± 7.5 | 101.4 | 11.1 | 13.0 | 10.1 | 6.7 | 8.9 |
| 2 | 5 | 8 | 4 | 2 | 8 | 4 | 2 | 865 ±36.6 | 109.9 | 12.7 | 13.7 | 14.6 | 6.3 | 5.8 |
| | 6 | 8 | 4 | 2 | 8 | 2 | 4 | 908 ±33.9 | 133.0 | 14.6 | 14.5 | 14.7 | 5.9 | 5.8 |
| | 7 | 8 | 4 | 2 | 9 | 1 | 4 | 918 ±23.0 | 133.6 | 14.6 | 14.5 | 14.6 | 5.9 | 5.9 |
| | 8 | 8 | 4 | 2 | 6 | 3 | 5 | 823 ±31.5 | 123.1 | 15.0 | 14.9 | 15.0 | 5.7 | 5.7 |
| 3 | 9 | 8 | 2 | 4 | 8 | 2 | 4 | 920 ±40.7 | 118.0 | 12.8 | 13.9 | 12.2 | 6.2 | 7.2 |
| | 10 | 8 | 2 | 4 | 8 | 4 | 2 | 877 ±22.7 | 124.9 | 14.2 | 13.2 | 14.9 | 6.6 | 5.7 |
| | 11 | 8 | 2 | 4 | 9 | 1 | 4 | 1001 ± 2.1 | 134.9 | 13.5 | 13.3 | 13.5 | 6.5 | 6.4 |
| | 12 | 8 | 2 | 4 | 6 | 3 | 5 | 855 ± 4.6 | 117.9 | 13.8 | 14.1 | 13.6 | 5.1 | 5.4 |
| 4 | 13 | 9 | 1 | 4 | 9 | 1 | 4 | 807 ± 8.2 | 131.7 | 14.5 | 13.8 | 14.9 | 6.3 | 5.8 |
| | 14 | 9 | 1 | 4 | 8 | 2 | 4 | 912 ± 9.7 | 147.5 | 16.2 | 15.4 | 16.4 | 5.5 | 5.1 |
| | 15 | 9 | 1 | 4 | 8 | 4 | 2 | 928 ±23.9 | 134.9 | 14.5 | 14.6 | 14.5 | 5.9 | 5.9 |
| | 16 | 9 | 1 | 4 | 6 | 3 | 5 | 800 ±23.9 | 130.4 | 16.3 | 15.3 | 17.1 | 5.5 | 4.9 |
| 5 | 1 | 6 | 3 | 5 | 6 | 3 | 5 | 765 ±32.5 | 109.1 | 14.4 | 14.1 | 14.6 | 6.1 | 5.9 |
| | 8 | 8 | 4 | 2 | 6 | 3 | 5 | 823 ±31.5 | 123.1 | 15.0 | 14.9 | 15.0 | 5.7 | 5.7 |
| | 12 | 8 | 2 | 4 | 6 | 3 | 5 | 855 ± 4.6 | 117.9 | 13.8 | 14.1 | 13.6 | 5.1 | 5.4 |
| | 16 | 9 | 1 | 4 | 6 | 3 | 5 | 800 ±23.9 | 130.4 | 16.3 | 15.3 | 17.1 | 5.5 | 4.9 |
| 6 | 2 | 6 | 3 | 5 | 8 | 4 | 2 | 722 ±17.8 | 102.0 | 13.2 | 14.6 | 12.0 | 5.9 | 7.3 |
| | 5 | 8 | 4 | 2 | 8 | 4 | 2 | 865 ±36.6 | 109.9 | 12.7 | 13.7 | 14.6 | 6.3 | 5.8 |
| | 10 | 8 | 2 | 4 | 8 | 4 | 2 | 877 ±22.7 | 124.9 | 14.2 | 13.2 | 14.9 | 6.6 | 5.7 |
| | 15 | 9 | 1 | 4 | 8 | 4 | 2 | 928 ±23.9 | 134.9 | 14.5 | 14.6 | 14.5 | 5.9 | 5.9 |
| 7 | 3 | 6 | 3 | 5 | 8 | 2 | 4 | 862 ±31.6 | 103.8 | 12.0 | 14.1 | 11.1 | 6.1 | 8.0 |
| | 6 | 8 | 4 | 2 | 8 | 2 | 4 | 908 ±33.9 | 133.0 | 14.6 | 14.5 | 14.7 | 5.9 | 5.8 |
| | 9 | 8 | 2 | 4 | 8 | 2 | 4 | 920 ±40.7 | 118.0 | 12.8 | 13.9 | 12.2 | 6.2 | 7.2 |
| | 14 | 9 | 1 | 4 | 8 | 2 | 4 | 912 ± 9.7 | 147.5 | 16.2 | 15.4 | 16.4 | 5.5 | 5.1 |
| 8 | 4 | 6 | 3 | 5 | 9 | 1 | 4 | 916 ± 7.5 | 101.4 | 11.1 | 13.0 | 10.1 | 6.7 | 8.9 |
| | 7 | 8 | 4 | 2 | 9 | 1 | 4 | 918 ±23.0 | 133.6 | 14.6 | 14.5 | 14.6 | 5.9 | 5.9 |
| | 11 | 8 | 2 | 4 | 9 | 1 | 4 | 1001 ± 2.1 | 134.9 | 13.5 | 13.3 | 13.6 | 6.5 | 6.4 |
| | 13 | 9 | 1 | 4 | 9 | 1 | 4 | 907 ± 8.2 | 131.7 | 14.5 | 13.8 | 14.9 | 6.3 | 5.8 |

* Difference between means necessary for significance at the 5% level = 45.7.

the increase in $SO_4$ or the decrease in $PO_4$ since, in accordance with the design of the experiment, as the concentration of $SO_4$ is increased the concentration of $PO_4$ is proportionately decreased. On the other hand, definite evidence that $SO_4$ was not toxic when used at its maximum value in this series is presented in sub-series 3 and 4. Here, the difference in yield between the anion ratios 8-2-4 and 8-4-2 was not statistically significant in either case. Thus, it is evident that the plant has a wide range of tolerance of $SO_4$, and a supply of this ion in the substrate ranging from 1 to 4 relative parts is favourable to growth.

A study of the data on the cations reveals that no individual cation ratio shows definite superiority throughout the series. However, it is evident that the yield produced by the 6-3-5 cation distribution was significantly lower than that of any other treatment in all sub-series except 8. In each sub-series, the highest yield was produced when 8 or 9 parts of the total cation concentration were supplies as Ca and 1 or 2 parts as Mg; however, it is not conclusive that the latter ion was toxic at any concentration used in this series. It is indicated that K is required at a relatively high concentration as yet not determined precisely.

As in Series I, high $NO_3$ and high K supply were each associated with high moisture content in the leaf tissue and high Ca supply was associated with high dry weight in the leaf tissue.

These data show that optimal growth can be produced when 1 to 4 parts of the anion concentration are supplied as $SO_4$ and 1 to 2 parts of the cation concentration as Mg. However, the most favourable concentration of each of these ions seems to be 2 parts since this amount is well above the deficiency level and any increase in the amount of either ion would necessitate a corresponding decrease in the amount of another ion of similar charge. The need of further investigation to establish the most favourable concentration of each of the other nutrient ions is indicated.

*Series III*

In this series, the total osmotic concentration of each nutrient solution was increased to 0.75 atmosphere. The ionic concentrations were adjusted in increments corresponding to multiples of the one-fifteenth increment of the total concentration of the anions and the cations respectively. The concentration of each of Mg and $SO_4$ was held constant at the 2-part level throughout the experiment and each of the other four major ions was used at the following levels: 6.5, and 8 parts. Nine solutions were devised as shown in Table 3, the ionic proportions employed being such that they could be made from 5 salts, namely $KH_2PO_4$, $Ca(NO_3)_2$, $MgSO_4$, $CaH_4(PO_4)_2$, and $KNO_3$. Six replications of each treatment were used. The seedlings were planted on October 2, 1940, and the plants were harvested on January 14, 1941.

The results of this series are given in Table 3. The data on the anions (sub-series 1 to 3) show that the yield produced by the treatment high in $NO_3$ was significantly higher than that of any other treatment at all cation proportions. No significance can be ascribed to the differences in yield between the other anion proportions except in sub-series 3 in which the 6.5-2-6.5 ratio ranked second. These results affirm the previous finding

TABLE 3.—YIELD DATA FOR SERIES III, ARRANGED IN SUB-SERIES 1–3 FOR COMPARISON OF ANION EFFECT UPON GROWTH AND IN SUB-SERIES 4–6 FOR COMPARISON OF CATION EFFECT UPON GROWTH

| Sub-series No. | Treatment No. | Cation proportions | | | Anion proportions | | | Mean fresh weight per plant | Mean dry weight per plant | Per cent dry weight | | | | Water per gram dry weight | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Ca | Mg | K | $NO_3$ | $SO_4$ | $PO_4$ | | | Per plant | Stalks | Top leaves | Bottom leaves | Stalks | Top leaves | Bottom leaves |
| | | | | | | | | gm. * | gm. | % | % | % | % | gm. | gm. | gm. |
| 1 | 1 | 5 | 2 | 8 | 5 | 2 | 8 | 924 ± 35.7 | 93.9 | 10.2 | 14.8 | 10.0 | 9.4 | 5.7 | 10.0 | 9.0 |
| | 2 | 5 | 2 | 8 | 6.5 | 2 | 6.5 | 995 ± 62.3 | 97.3 | 9.8 | 10.8 | 10.1 | 9.2 | 8.7 | 8.8 | 9.8 |
| | 3 | 5 | 2 | 8 | 8 | 2 | 5 | 1157 ± 86.9 | 113.3 | 9.8 | 12.0 | 10.0 | 7.4 | 7.3 | 10.0 | 12.5 |
| 2 | 4 | 6.5 | 2 | 6.5 | 5 | 2 | 8 | 918 ± 50.4 | 115.6 | 11.8 | 15.2 | 13.0 | 7.6 | 5.5 | 6.6 | 12.1 |
| | 5 | 6.5 | 2 | 6.5 | 6.5 | 2 | 6.5 | 984 ± 27.3 | 141.3 | 14.4 | 15.0 | 13.0 | 14.6 | 5.6 | 6.6 | 5.7 |
| | 6 | 6.5 | 2 | 6.5 | 8 | 2 | 5 | 1097 ± 31.9 | 129.3 | 11.8 | 13.0 | 14.4 | 8.2 | 6.6 | 5.9 | 11.1 |
| 3 | 7 | 8 | 2 | 5 | 5 | 2 | 8 | 937 ± 53.2 | 105.0 | 10.6 | 12.0 | 12.2 | 9.6 | 7.3 | 7.2 | 9.4 |
| | 8 | 8 | 2 | 5 | 6.5 | 2 | 6.5 | 1037 ± 52.8 | 127.1 | 12.3 | 14.0 | 12.8 | 10.0 | 6.1 | 6.8 | 9.0 |
| | 9 | 8 | 2 | 5 | 8 | 2 | 5 | 1150 ±102.5 | 132.0 | 11.5 | 13.4 | 11.8 | 6.2 | 6.5 | 7.4 | 15.1 |
| 4 | 1 | 5 | 2 | 8 | 5 | 2 | 8 | 924 ± 35.7 | 93.9 | 10.2 | 14.8 | 10.0 | 9.4 | 5.7 | 10.0 | 9.0 |
| | 4 | 6.5 | 2 | 6.5 | 5 | 2 | 8 | 918 ± 50.4 | 115.6 | 11.8 | 15.2 | 13.0 | 7.6 | 5.5 | 6.6 | 12.1 |
| | 7 | 8 | 2 | 5 | 5 | 2 | 8 | 937 ± 53.2 | 105.0 | 10.6 | 12.0 | 12.2 | 9.6 | 7.3 | 7.2 | 9.4 |
| 5 | 2 | 5 | 2 | 8 | 6.5 | 2 | 6.5 | 995 ± 62.3 | 97.3 | 9.8 | 10.8 | 10.1 | 9.2 | 8.7 | 8.8 | 9.8 |
| | 5 | 6.5 | 2 | 6.5 | 6.5 | 2 | 6.5 | 984 ± 27.3 | 141.3 | 14.4 | 15.0 | 13.0 | 14.6 | 5.6 | 6.6 | 5.7 |
| | 8 | 8 | 2 | 5 | 6.5 | 2 | 6.5 | 1037 ± 52.8 | 127.1 | 12.3 | 14.0 | 12.8 | 10.0 | 6.1 | 6.8 | 9.0 |
| 6 | 3 | 5 | 2 | 8 | 8 | 2 | 5 | 1157 ± 86.9 | 113.3 | 9.8 | 12.0 | 10.0 | 7.4 | 7.3 | 10.0 | 12.5 |
| | 6 | 6.5 | 2 | 6.5 | 8 | 2 | 5 | 1097 ± 31.9 | 129.3 | 11.8 | 13.0 | 14.4 | 8.2 | 6.6 | 5.9 | 11.1 |
| | 9 | 8 | 2 | 5 | 8 | 2 | 5 | 1150 ±102.5 | 132.0 | 11.5 | 13.4 | 11.8 | 6.2 | 6.5 | 7.4 | 15.1 |

* Difference between means necessary for significance at the 1% level = 73.7.

that $NO_3$ is required at a high concentration and indicate that an amount of $PO_4$ higher than 5 relative parts of the anion concentration is in excess of the requirement of the plant for maximum growth.

The differences in yield associated with variations in the cation proportions (sub-series 4 to 6) are not significant at any of the three anion proportions. This shows that the variations in the cation proportions employed in this series do not result in a significant variation in the yield provided the positively charged ions are at an optimum level. Therefore, a concentration of 5 to 8 parts of each of Ca and K is adequate for best growth.

The variations in ionic concentrations in this series were not sufficiently great to provide a basis for the study of the influence of each ion on the dry weight percentage of the tissue.

## GENERAL DISCUSSION

It is evident that $NO_3$ is the anion which has the greatest influence over growth under the conditions of these experiments and that this influence is correlated with the relative cation levels. An increase in the mean fresh weight deviation indicates an increase in the importance of the cation ratio. In series I, the mean fresh weight deviation in sub-series 5, 6, and 7 was 8, 8.8 and 27.2 grams respectively. A comparison of these results shows that the efficient utilization by the plant of high $NO_3$ supply is greatly influenced by the relative cation supply. Thus, at low $NO_3$ levels (sub-series 5 and 6) the influence of variations in the cation distribution was slight and the yield was affected very little; but, at the highest $NO_3$ level (sub-series 7), the effectiveness of this ion in producing high yield was conditioned to a greater degree by the relative cation concentrations. Beckenback *et al.* (1) reported similarly for corn.

The $PO_4$ ranks second in importance among the anions and the $SO_4$ third. There was no indication of a deficiency of $SO_4$ at the lowest level and the evidence is not conclusive that this ion was toxic at any of the concentrations tested. The depression of growth in series II when $SO_4$ was used at a higher concentration than that of $PO_4$ may be attributable to a decrease in the supply of $PO_4$, rather than to toxicity of $SO_4$ since evidence of toxicity of the latter when used at the higher levels in series I was lacking in all cases except sub-series 4. Therefore, the optimum range of $SO_4$ is quite wide.

Of the cations, it is shown that the solution required for maximum growth contains 5 to 8 relative parts of each of Ca and K, the combined concentration of these two ions being 13 parts out of a total of 15 relative parts of the cation concentration. The remaining 2 parts are supplied as Mg. The maximum concentration of Mg in conjunction with low Ca and K resulted in a reduction in yield and in the appearance of necrotic areas on the leaves. This necrosis was reduced in severity when the anion ratio was high in $PO_4$ and low in $NO_3$ and $SO_4$.

An examination of the data leads to the conclusion that no exact ionic ratio is essential for optimum nutrition. The plant's power of adaptation seems to enable it to respond favourably to considerable variations in the concentrations of nutrient ions. Good growth was produced over a wide range of ionic ratios, provided the total supply of essential elements

was adequate. It is clearly indicated that, while there is no specific requirement of each ion for optimum nutrition, there is a definite range of concentrations of each ion for optimum requirements.

Based on the results of these investigations, the concentration range of each of the 6 major nutrient ions for optimum nutrition of White Mammoth is given in Section A of Table 4 in terms of the relative parts of the concentration of the ions as used in this work. The data in Section B of Table 4 give the concentration of the nutrient element contained in each ion per million parts of the nutrient solution at the ionic levels given in Section A.

TABLE 4

| A—THE CONCENTRATION RANGE OF THE SIX MAJOR NUTRIENT IONS FOR OPTIMUM NUTRITION OF WHITE MAMMOTH | | B—THE CONCENTRATION IN P.P.M. OF THE NUTRIENT ELEMENT CONTAINED IN EACH MAJOR ION AT THE IONIC LEVELS GIVEN IN TABLE 4A | |
|---|---|---|---|
| Nutrient ion | Relative parts of concentration of ions | Nutrient element | P.p.m. of nutrient element |
| Ca | 5 – 8 | Ca | 150 – 240 |
| Mg | 1 – 2 | Mg | 34 – 68 |
| K | 5 – 8 | K | 200 – 320 |
| $NO_3$ | 8 – 9 | N | 168 – 189 |
| $SO_4$ | 1 – 4 | S | 45 – 180 |
| $PO_4$ | 4 – 5 | P | 124 – 155 |

Further physiological effects of the inidvidual ions are revealed by the dry weight data. It is shown that the concentration of $NO_3$ in the nutrient medium is the primary factor in determining the proportion of dry weight. An increase in the $NO_3$ supply results in an increase in the proportion of water in the leaf tissue, while the relative proportions of $SO_4$ and $PO_4$ do not seem to have any definite influence on the moisture content of the tissue. Of the cations, a high level of K in the nutrient solution produced plants with a relatively high percentage of water in the leaf tissue, while a high level of Ca in the solution resulted in a high percentage of dry matter in this tissue.

## SUMMARY

A study was made of the effect of the six major nutrient ions on the growth of flue-cured tobacco in artificial culture and an optimum nutrient solution was developed for the variety White Mammoth. It was shown that no exact ionic ratio in the nutrient medium is required for maximum growth, but the optimum concentration range of each nutrient ion was determined.

The highest correlation between the nutrient supply and the growth of the plant was observed for $NO_3$ between the limits of one and nine, out of a total of 15 relative parts of the anion concentration. $NO_3$ was followed in order of influence of the anions on growth by $PO_4$ and $SO_4$. The plant manifested a wide range of tolerance of $SO_4$.

At increasingly high concentrations of $NO_3$ in the nutrient solution, the effectiveness of this ion in producing high yield was conditioned in increasing degree by the relative proportions of Ca, Mg, and K in the nutrient solution. At high $NO_3$ levels, greatest growth was produced when Ca and K were each supplied at a concentration of 5 to 8 parts out of a total of 15 relative parts of the cation concentration, the combined concentration of the two ions being 13 parts. The remaining 2 parts were supplied as Mg.

An increase in the nutrient supply of each of $NO_3$ and K was associated with an increase in the percentage of water in the leaf tissue while an increase in the supply of Ca was associated with an increase in the percentage of dry material in the leaf tissue. No relationship was observed between the supply of $PO_4$, $SO_4$, and Mg and the relative proportions of water and dry matter in the tissue.

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# A NOTE ON THE PRODUCTION OF VITAMIN C BY SPROUTING SEEDS[1]

L. P. V. Johnson[2], G. A. Young,[3] and J. B. Marshall[2]

*National Research Laboratories, Ottawa*

The antiscorbutic value of germinated seeds has been known since the time of Captain Cook, and was investigated in Norway prior to the last war (5). After the Mesopotamian campaign of 1915–16 British workers directed attention to the possibility of utilizing this property as a source of accessory dietary factors for troops separated from supplies of fresh food and for the relief of famine stricken areas (1). Advances since 1918 in the knowledge of the chemistry of vitamins have made it possible to investigate more comprehensively methods for the production and preparation of an antiscorbutic material from sprouted seeds. In this note the results are given of a survey to determine the suitability, for this purpose, of seeds readily available in Canada, as well as some others less commonly known.

Legumes have been considered more prolific producers of ascorbic acid than cereals (5) and there is evidence of physiological differences between crop varieties as well. Bonner and Bonner (3) showed that pea varieties could be grouped according to their response to the presence of ascorbic acid in the media in which excised embryos were cultured. This response was related inversely to the concentration of ascorbic acid initially present in the embryos. The presence of ascorbic acid in actively growing tissues, i.e. hypocotyl, and later in chlorophyll bearing tissues has been demonstrated by Reid (4).

## MATERIALS

The material examined included varieties of the following crops: common and garden peas, *Pisum sativum;* common bean, *Phaseolus vulgaris;* green gram bean[4], *P. aureus*; lima bean, *P. lunatus*; English broad bean, *Vicia fabia;* soy bean, *Glycine soja*; spring wheat, *Triticum sativum*; hull-less oats, *Avena nuda*; corn, *Zea mays, v. saccharata*; okra, *Hibiscus esculentus.* A complete list including variety names is given in Table 1. The seed used in each case was of good quality, having been graded previously for seed purposes. This initial quality is of importance as sound viable seed gives some assurance of optimum performance in the sprouting tests. Broken and disease infected seeds tend to decompose readily and this may render the odour of an entire batch of seed offensive.

## METHODS

Preliminary tests of the above material were made by germinating the seeds in petri dishes at 22.2° C. (72° F.) after soaking for 24 hours in tap

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[2] Biologist.

[3] Laboratory Steward.

[4] This species is closely related to the mung or black gram bean, *Phaseolus mungo* (2).

TABLE 1.—VARIETIES USED IN PRELIMINARY TESTS OF ASCORBIC ACID PRODUCTION

*Common pea, Pisum sativum*
- Field varieties
  - Arthur
  - Chancellor
  - Dashaway
  - Early Blue (Ottawa No. 21)
  - Early Britain
  - Golden Vine (1)*
  - Mackay (Ottawa No. 25) (1)
  - New Canadian Beauty
  - Potter
  - Prussian Blue (1)
- Garden varieties
  - Dwarf Gray Sugar
  - Little Marvel
  - Prince of Wales (1)
  - Thomas Laxton

*Soy bean, Glycine soja*
- Blackeye (1) (2)
- Mandarin

*Okra, Hibiscus esculentus*
- White Velvet (3)

*Common bean, Phaseolus vulgaris*
- Common white (1)
- Marrow fat (1)
- Red Kidney (1)

*Lima Bean, P. lunatus*
- Burpee Bush Lima (1)
- Henderson Bush Lima (1)

*Green Gram bean, P. aureus*
- Chinese Salad

*English Broad bean, Vicia fabia*
- Broad Windsor

*Spring wheat, Triticum sativum*
- Marquis
- Regent (2)
- Thatcher (2)

*Hull-less oats, Avena nuda*
- Laurel

*Corn (maize), Zea mays v. saccharata*
- Golden Bantam

* Reasons for elimination from further tests:
(1) Soured during germination.
(2) Low vitamin C production
(3) Poor germination.

water. Ascorbic acid determinations were made at daily intervals, commencing when the sprouts first appeared and continuing until the assay values began to decline. A number of varieties were eliminated on this basis.

The material thus selected was tested at three temperatures, 15.6°, 22.2°, and 28.9° C. (60°, 72°, and 84° F.) and by two methods of germination. In one, the seeds were soaked in trays for 24 hours, drained, spread to a depth of $\frac{3}{4}$ inch and flushed with water at the temperature required. The other method was similar to that recommended for use by the Australian Forces (6); seed was placed in loose cheese cloth bags, soaked 24 hours and thereafter dipped 4 times daily in water held at the germination temperatures.

Vitamin C assays were made on samples of the entire sprouted material. No attempt was made to separate the sprouts from the seeds and seed coats. Reduced ascorbic acid was determined by the dye titration method, 2% metaphosphoric acid being used to extract the material in a *Waring Blendor*. Moisture determinations were made on the material as well, in order to allow for the diluting effect of the tissue water. The results are expressed as mg. ascorbic acid per 100 gm. of sprouted material.

## RESULTS

In the preliminary tests 14 of the 31 varieties listed in Table 1 were eliminated for various reasons. A number of the pea and bean varieties

showed a pronounced tendency to become sour and offensive when germinated in this manner. Two of the wheat varieties were dropped because of low vitamin C production, and tests of okra were discontinued because of poor germination of the seed used.

For convenience and expediency the bag method of germination was used for most of the tests, and the data presented are based on this method. The two methods did not differ appreciably. Considerable care was required with the trays that were used, to avoid souring as a result of impeded drainage and lack of aeration. Trays with perforated bottoms would have been much more satisfactory.

Data showing the effects of temperature on vitamin C production and the condition of the sprouted material are given in Table 2. Rapid souring or spoilage of all the material except the Chinese Salad bean resulted at 28.9° C. Of the other temperatures the lower (15.6° C.) was more favourable for the legumes and the higher (22.2° C.) for the cereals.

TABLE 2.—EFFECT OF TEMPERATURE ON THE VITAMIN C PRODUCTION AND GROWTH OF SEEDS OF SELECTED VARIETIES GERMINATED IN BAGS

| Crop | Variety | Temp. °C. | No. days for maximum production | *Growth | Condition of material | Mg./100 gm. of material | | Percentage moisture |
|---|---|---|---|---|---|---|---|---|
| | | | | | | Wet | Dry | |
| Bean | Chinese Salad | 15.6 | 3 | Good | Fresh | 23 | 89 | 74 |
| | English Broad Windsor | | 7 | Very good | Slightly sour | 33 | 122 | 73 |
| | Mandarin Soy | | 4 | Fair | Fresh | 4 | 13 | 68 |
| Corn | Golden Bantam | | 7 | Good | Sour | 7 | 20 | 63 |
| Oats | Laurel | | 6 | Very good | Fresh | 8 | 30 | 75 |
| Pea | Arthur | | 5 | Very good | Fresh | 17 | 61 | 67 |
| Wheat | Marquis | | 5 | Very good | Fresh | 4 | 16 | 75 |
| Bean | Chinese Salad | 22.2 | 2 | Very good | Fresh | 21 | 65 | 68 |
| | English Broad Windsor | | 4 | Good | Sour | 23 | 73 | 69 |
| | Mandarin Soy. | | 3 | Poor | Sour | 2 | 3 | 32 |
| Corn | Golden Bantam | | 4 | Very good | Fresh | 10 | 30 | 65 |
| Oats | Laurel | | 6 | Very good | Slightly sour | 11 | 19 | 43 |
| Pea | Arthur | | 3 | Fair | Fresh | 3 | 7 | 64 |
| Wheat | Marquis | | 4 | Very good | | 5 | 13 | 65 |
| Bean | Chinese Salad | 28.9 | 2 | Fair | Fresh | 18 | 64 | 73 |
| | English Broad Windsor | | 2 | Poor | Sour | — | — | — |
| | Mandarin Soy | | 2 | Poor | Sour | — | — | — |
| Corn | Golden Bantam | | 2 | Poor | Sour | — | — | — |
| Oats | Laurel | | 2 | Poor | Sour | 1 | 1 | 43 |
| Pea | Arthur | | 2 | Poor | Sour | — | — | — |
| Wheat | Marquis | | 2 | Fair | Slightly sour | 1 | 2 | 47 |

* Growth poor, less than 50% germination; fair, less than 75% germination; good, less than 95% germination; very good, 95 to 100% germination.

A comparison of 12 varieties of peas sprouted in bags at 15.6° C. is given in Table 3. Early Blue (Ottawa No. 21), O.A.C. No. 181 and Arthur

TABLE 3.—VITAMIN C PRODUCTION AND GROWTH IN FIELD AND GARDEN PEAS, SPROUTED IN BAGS AT 15.6° C.

| Variety | No. days for maximum production | Growth | Mg./100 gm. of material wet basis |
|---|---|---|---|
| Arthur | 5 | Good | 17 |
| Chancellor | 4 | Poor | 8 |
| Dwarf Grey Sugar | 4 | Fair | 11 |
| Dashaway | 4 | Fair | 12 |
| Early Blue, Ottawa No. 21 | 5 | Good | 20 |
| Early Britain | 4 | Fair | 11 |
| Little Marvel | 4 | Fair | 14 |
| New Canadian Beauty | 4 | Poor | 10 |
| O.A.C. No. 181 | 5 | Good | 18 |
| Potter | 4 | Good | 13 |
| Thomas Laxton | 4 | Good | 10 |
| White Wonder | 4 | Fair | 9 |

took 5 days to reach their maximum vitamin C production (17 to 20 mg. per 100 g.), while the other varieties began to decline at 4 days, after having reached a maximum of 8 to 14 mg. per 100 g.

In Figure 1 the daily increases in the ascorbic acid content of 100 gm. of sprouting material are shown for selected seeds, germinated in bags at 15.6° C. The Chinese Salad bean reached a maximum in 3 days while the Broad Windsor bean took 7 days and the Early Blue pea 5 days. Mandarin soy bean and Laurel oats did not approach the level of the others but were included for comparison.

## CONCLUSIONS

Of the seeds tested in the present investigation, the Chinese Salad bean appeared to have the greatest range of adaptability combined with rapid production of vitamin C. However it is not available in sufficient quantities to warrant its recommendation for use on a large scale. The field pea varieties, Early Blue (Ottawa No. 21), O.A.C. No. 181, and Arthur produced appreciable amounts of antiscorbutic in 5 days and are readily available. The English Windsor broad bean is in less favourable supply but showed considerable merit with respect to vitamin C production, particularly at the lowest temperature.

The method of germination to be employed will be determined by circumstances and should be investigated in greater detail. Experience gained from the present experiments indicated the desirability of temperatures between 15 and 22° C. (60° and 72° F.) and of importance of drainage and aeration for the germinating material.

## ACKNOWLEDGMENTS

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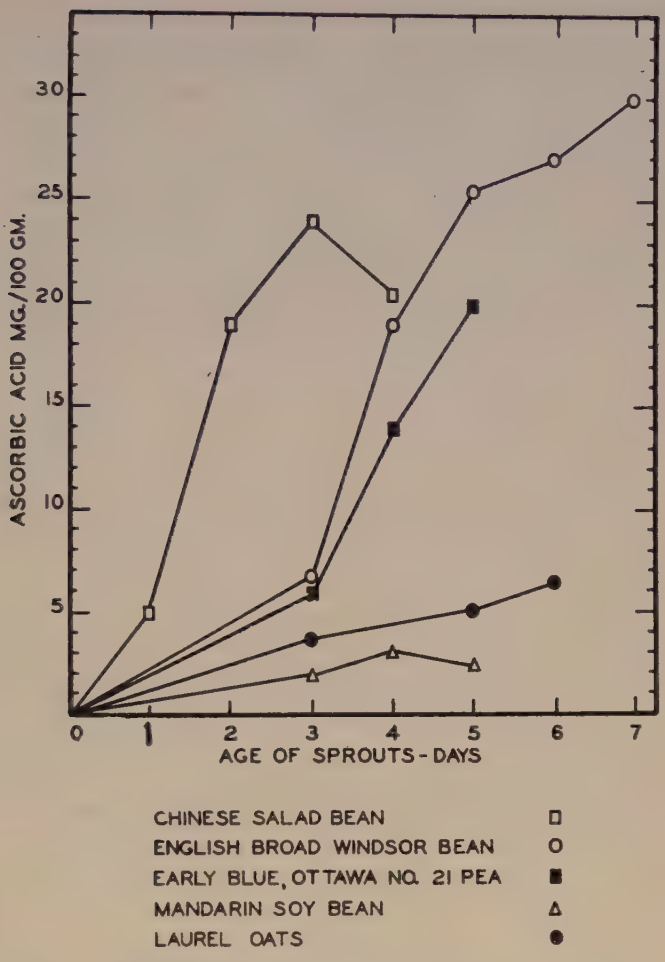


Figure 1.—Vitamin C production of selected seeds at 15.6°C. (60°F.)

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# LEGUME AND CEREAL SPROUTS AS A DIETARY SUBSTITUTE FOR FRESH VEGETABLES[1]

W. A. Andreae[2], Edith A. Chalmers[2], and W. D. McFarlane[3]

*Macdonald College, (McGill University) Quebec*

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Cereal and legume sprouts have been used as food for many centuries in the Orient, and records also exist which show that the early explorers cured or prevented scurvy by eating sprouted peas and beans when fresh vegetables were not available. Soybean sprouts have recently received much publicity in the United States as the result of investigations by Professor C. M. McCay of Cornell University.

Dry seeds, in general, are devoid of ascorbic acid, but the seedlings contain a considerable amount. Burkholder (1) reports an appreciable increase in riboflavin and other members of the vitamin B-complex during the growth of corn, wheat, oats and barley seedlings. Sprouts have a flavour and nutritive value which approximates that of fresh vegetables and in addition, possess several advantages in that the seeds can be sprouted, ready for use in less than 10 days and their cultivation is not limited by season, climate, sunlight or soil. The only requirements are a fairly constant room temperature of about 70° F. and a good water supply.

It might be possible to supply a significant part of the daily requirements for ascorbic acid and vitamin B-complex by the use of sprouts in the diet and hence contribute to the solution of nutritional problems arising out of the difficulty in transporting fresh vegetables to troops in isolated areas and to the population of countries released from enemy occupation. Fresh vegetables are perishable and bulky and require rapid transportation and adequate storage facilities, whereas dry seeds may be shipped to the desired location and sprouted at their destination.

The primary aims of the investigations recorded below were *(a)* to devise simple and inexpensive methods for producing sprouts in bulk, even under adverse conditions; *(b)* determine the best time to harvest for highest vitamin content commensurate with palatability; and *(c)* prepare appetizing dishes containing sprouts.

## MATERIALS AND METHODS

### *Sprouting*

Good sprouts can only be obtained from good seed; broken seeds must be eliminated as they will constitute a centre for infection. Disinfectants, such as bleaching powder, at the low concentration of 1 tablespoon to 4 gallons of water, will destroy surface bacteria without harming the seed but will not prevent all bacterial growth; equally effective results were obtained with an abundance of fresh water. The sprouting was

[1] Macdonald College Journal Series No. 198.
[2] Graduate Research Assistants, Department of Chemistry, Faculty of Agriculture.
[3] Professor of Chemistry.

carried out in a basement room with two small windows, a well-drained cement floor, and a good water supply. There was no provision for control of the room temperature which fluctuated from 66 to 73° F.

The seeds were soaked for 24 hours and held at a high humidity and a temperature of about 22° C. The most suitable container was found to be a 6-gallon lard tub with small holes in the bottom for drainage. The tub was half-filled with soaked seeds and covered with a lid to exclude light and to maintain a high humidity inside. The tub was filled twice daily with water and the seeds stirred by hand to break up the sprouting mass. The water drained through the holes in the bottom, leaving the sprouts moistened, washed and cooled. For smaller quantities, wide-mouthed fruit jars were used. The soaked seeds were placed in the jar and a piece of cheese-cloth was tied over the opening. The jar was filled with water twice daily, inverted to drain and allowed to remain in this position until the next watering.

These methods were not suitable for the cultivation of green shoots. In this case the soaked seeds were spread thinly on flat wire-screen trays, 14″ × 22″, supported at each corner by wooden legs, 7″ high. These trays (Figure 1) are light and easy to handle and can be stacked to facilitate

FIGURE 1. Tray method of cultivating green shoots.

watering and utilize space. A layer of wood shavings on the screen increased the moisture retention and facilitated the cleaning of the trays after the shoots had been cut.

A portable container suitable for sprouting all types of seeds was made from a piece of coarse jute 40" × 36" (Figure 2). The bottom of this bag measured 15" × 19" and the sides were 6" high. Four sticks, less than 1" in diameter, were pushed through the open hems around the bottom, giving the bag considerable rigidity. Each corner was provided with a sling by

FIGURE 2. Portable container for sprouting all types of seeds

which the bag could be suspended by two poles or hooked to some convenient device. The bag was rendered rot proof by first immersing it in a 2% copper sulphate solution, then in 5% sodium carbonate solution and finally rinsing well with water. Rot proofing had no detrimental effect on the sprouting.

The seeds used were selected on the basis of *(a)* availability in Canada, *(b)* ease of sprouting, *(c)* nutritive value of the sprouts and *(d)* palatability of the cooked product.

*Cooking*

The authors considered as valuable any sprouts which were palatable and would serve to break the monotony of a diet depleted in fresh vegetables. However, such a diet is also likely to be low in vitamin C so the authors preferred to utilize sprouts of a high ascorbic acid content. Meals which included sprouts were prepared and served in the Macdonald College dining room. The final judgments of the palatability of the cooked food were obtained from C.W.A.C. personnel and College students, through the medium of a questionnaire.

The simplest cooking methods were employed. In boiling, the water was always brought to the boiling point, the sprouted seeds added, the container covered tightly and its contents brought back to boiling temperature as rapidly as possible. A minimum amount of water was used, but in cooking sprouted broad-beans they had to be almost covered with water to provide uniform cooking throughout. To preserve vitamin C emphasis was placed on preparing dishes which did not entail loss in the cooking water, e.g., soup, stew, baked dishes and fried dishes. The cooking water was saved for use in soup stocks, gravies or sauces.

*Chemical Analysis*

*A. Ascorbic Acid*

The determination of reduced ascorbic acid was carried out as follows:

Two 10-g. samples were ground with acid-washed sand and 25 ml. of normal-sulfuric acid containing 2% metaphosphoric acid. The extracts were centrifuged and washed three times with 20-ml. portions of the acid mixture. The combined extracts were made up to 100 ml. and aliquots were titrated against a standard solution of 2–6 dichlorophenol indophenol, as outlined by Burrell and Ebright (2). To analyze bean sprouts duplicate 25-g. samples were weighed out and transferred to a Waring Blendor with 100 ml. of the acid and reduced to a pulp within 8 minutes. The extracts were then treated as described above but the centrifugate and washings were made up to 200 ml. This method permitted the use of larger and more representative samples as each seedling weighed about 5 g.

When turbid or coloured extracts were encountered, making it difficult to detect the end point, 10 ml. of chloroform were well mixed with an aliquot of the extract and the titration completed, according to McHenry and Graham (3). On centrifuging, two layers were formed and the end point could be determined in the chloroform layer.

Total ascorbic acid determinations (reduced plus dehydroascorbic acid) were made on some of the samples, especially cooked products, employing $H_2S$ reduction. The common procedure is to bubble $H_2S$ through the extract for 15 min. and remove the $H_2S$ with a stream of carbon dioxide or nitrogen. At least 2 hours are required to remove the $H_2S$, and frequently traces remain which reduce the indophenol dye and give results which are

too high. To completely remove the $H_2S$ in a shorter time Fellenberg (4) heated the extract and Harris and Olliver (5) put the extract under reduced pressure. The authors found the following procedure, using the apparatus illustrated in Figure 3 to be most satisfactory.

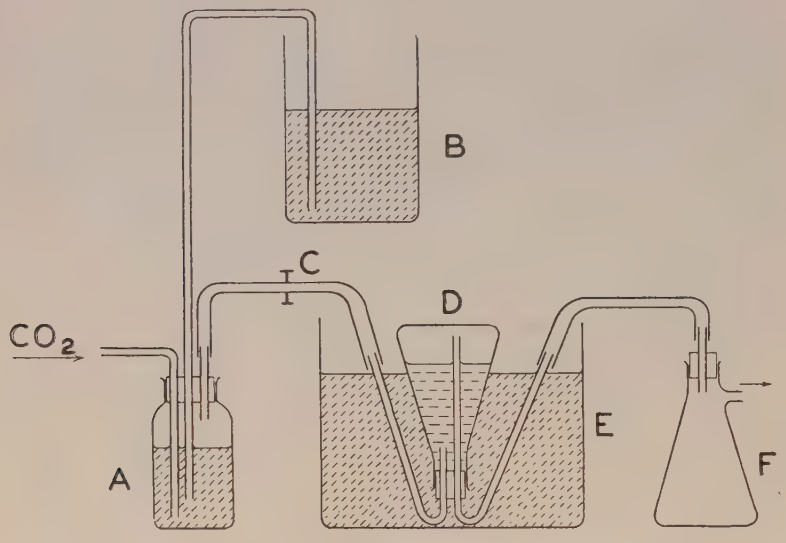


FIGURE 3. Apparatus for $H_2S$ reduction of dehydroascorbic acid.

A 50-ml. aliquot of the extract, adjusted to pH 3.0, was transferred to a 125-ml. Erlenmeyer flask (D) fitted with a two-hole stopper, an aeration tube with head close to the stopper, and a glass tube extending to the bottom of the flask. The flask was inverted in a water bath (E) at 55° C., and the $H_2S$ entered through the aeration tube and escaped at the top by the long glass tube. This device ensured complete saturation of the solution with $H_2S$ and prevented loss due to foaming. The reduction of dehydroascorbic acid was complete in 15 min. and the extract was then adjusted to pH 2.0 with 50% $H_2SO_4$. The flask was replaced in the water bath and connected to a large bottle (A). $CO_2$ was admitted into A which was filled with water and the gas displaced the water into the elevated bottle (B). A pinch-cock (C) controlled the gas flow to D and the whole system was put under a reduced pressure of 100 mm. Hg. by applying suction to flask (F). In this apparatus the extract was freed from $H_2S$ in 15 minutes at 55° C.

It is well known that the indophenol method is affected by the presence of other reducing substances in the extracts. Roe's colorimetric method (6, 7) employing 2,4 dinitrophenylhydrazine is said to be more specific and also determines dehydroascorbic acid. These methods were compared with the results shown in Table 1. In general the results agreed very well and did not indicate, so far as these samples were concerned, that any advantage would accrue from using the more complicated colorimetric procedure in place of the simple titration method.

TABLE 1.—ASCORBIC ACID DETERMINATIONS BY INDOPHENOL TITRATION AND BY THE COLORIMETRIC 2,4 DINITROPHENYLHYDRAZINE METHOD

| | Ascorbic acid—mg. per 100 g. | | | |
|---|---|---|---|---|
| | Reduced | | Total (reduced+dehydro.) | |
| | Titration | Colorimetric | Titration | Colorimetric |
| Broad Beans—sprouted 4 days | 24.6 | 22.9 | — | 29.2 |
| Vetch—sprouted 3 days | 12.1 | 12.8 | — | — |
| Vetch (cooked 20 min.) | 3.5 | 4.2 | — | 13.3 |
| Peas—sprouted 10 days | 20.5 | 19.1 | 21.9 | 24.5 |
| Dehydrated Cereal Grass | 41.2 | 43.5 | — | 45.7 |
| Standard Ascorbic Acid Solution (50 mg. per 100 cc.) | 50.00 | 49.8 | 50.0 | 50.3 |

*B. Riboflavin*

The riboflavin content of the sprouts was determined by the following method which combined certain features of the Chapman and McFarlane (8) and the Najjar (9) fluorimetric methods.

A 10-g. sample was weighed into a 250-ml. centrifuge bottle, 50 to 75 ml. of a 0.2% pepsin solution in 0.33% HCl were added and the mixture incubated overnight at 37° C. and in the dark. After adjusting to pH 4.5, using bromcresol-green as an external indicator, 0.1 g. of takadiastase was added and the incubation continued for 3 to 4 hours. The material was centrifuged, the supernatant transferred to a 100-ml. volumetric flask, the residue was twice washed with distilled water and the combined extracts made up to 100 ml. with distilled water.

5-ml. aliquots were shaken in a separatory funnel with 1 ml. glacial acetic acid and 2 ml. pyridine; 10 drops 4% potassium permanganate were added and after 1 minute 10 drops of 3% hydrogen peroxide. Finally, 5 g. anhydrous sodium sulphate and 10 ml. butyl alcohol were added, the mixture shaken thoroughly for 2 min. and centrifuged. The fluorescence of the pyridine-butyl alcohol phase was measured in a Coleman photofluorimeter. The fluorescence due to substances other than riboflavin was corrected for with a "blank" prepared by exposing the pyridine-butyl alcohol solution to intense ultra-violet radiations, for one hour, thus destroying riboflavin only. The fluorimeter was standardized by carrying out a determination with 5 ml. of a riboflavin solution containing one microgram of pure riboflavin.

## EXPERIMENTAL

### CEREALS

Cereal seeds are the most desirable from the standpoint of supply and low cost, but oats and barley have their seeds embedded in a coarse hull and are therefore not suitable for sprouting. Hulless oats and barley are now available, but the seed is still expensive and only small quantities are on the market. However, these were studied, along with rye and it was found that, after 3 days of sprouting, the optimum ascorbic acid content was 5–7 mg. per 100 gms. We investigated 5 common wheat varieties:

Red Bog, Thatcher, Apex, Regent and Marquis. When sprouted for 4 days, the ascorbic acid content remained low (1.5–2.5 mg. per 100 gms.) and reached a maximum of 5.5 mg. per 100 gms. at 10 days (Figure 4).

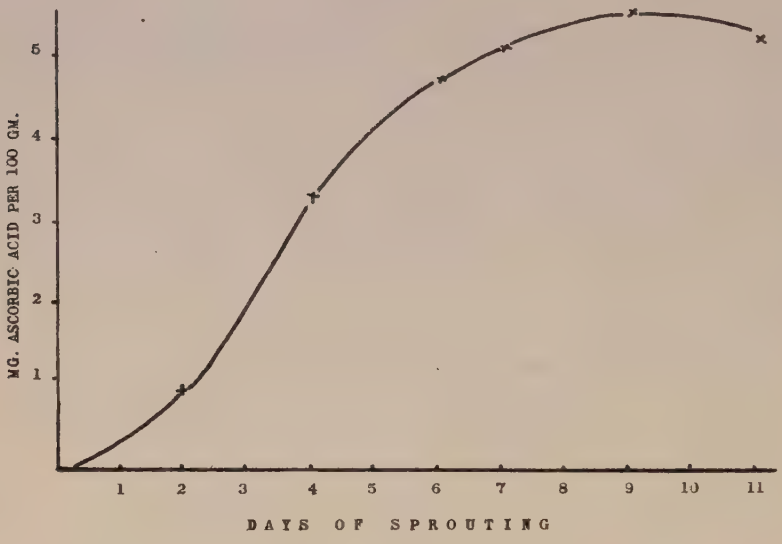


FIGURE 4. Showing the increase in ascorbic acid content of "Regent" wheat, during sprouting.

Cereal sprouts must be considered a poor source of ascorbic acid as 20–30% was lost on cooking for 10 minutes. Cereal sprouts alone were tasteless and had to be blended with oatmeal and served as porridge. It was found that after 3 days sprouting the roots were fibrous and unsuitable for human consumption, so that the sprouts could not be used at the stage of optimum vitamin C content. An attempt was made to remove the fibrous roots but the only practical method was to dry the sprouts, so that the roots became brittle and broke off on subsequent screening; this treatment destroyed the fresh food character of the sprouts.

## LEGUMES

Legume seeds do not develop fibrous roots during the early stage of sprouting, and the entire sprout can be eaten up to the stage of the appearance of green leaves. At more advanced stages, only the green shoots are edible. Sprouted Mung beans have been used primarily for dietary purposes but the beans cannot be grown in Canada. Soybean sprouts have attained some popularity, particular emphasis being laid on the protein and ascorbic acid content. The ascorbic acid content of the soybean varieties we have studied was low, amounting to 10–15 mg. per 100 g. after 5 days sprouting. About 30% of the ascorbic acid was concentrated in the tender shoots and was readily leached into the cooking water. The loss after 20 min. cooking was about 50%, thus reducing the ascorbic acid retention to such a low value,that their value was questionable. Since a

great deal of information was already available on sprouting soybeans, we concentrated our efforts on other seeds.

*1. Peas*

The C. S. I. R. Nutrition Laboratory, University of Adelaide, Australia, recommended the use of Blue Boiler peas, sprouted for 4 days. They claimed a vitamin C content of 38 mg. per 100 g., of which one-third was lost on cooking. This variety is not available in Canada. It was found that, with the variety Ottawa 181, the ascorbic acid content increased to 15–17 mg. per 100 g., after 5 days sprouting. Because of the hardness of the seeds, it was necessary to cook the sprouts for 1 hour to render them soft enough to be palatable, with the result that 80% of the vitamin was destroyed.

To reduce this loss, field peas were sprouted for 10 days, during which time the seeds sent up shoots 8″ long, so that the shoots could be clipped off close to the seeds and served as a green salad. This salad was agreeable to the taste and contained a significant amount of vitamin C. No advantage was found in prolonging growth beyond 10 days, as the shoots became fibrous and the ascorbic acid content declined. The distribution of ascorbic acid between the shoots, cotyledons and roots of pea seedlings at different stages of growth is shown in Figure 5. These sprouts were grown in a constant temperature chamber at 28° C. and 50% relative humidity. The sprouts grew rather poorly because of the low humidity. For comparison, the ascorbic acid content of seedlings from the same seed grown at 90% relative humidity, is also given in Figure 5.

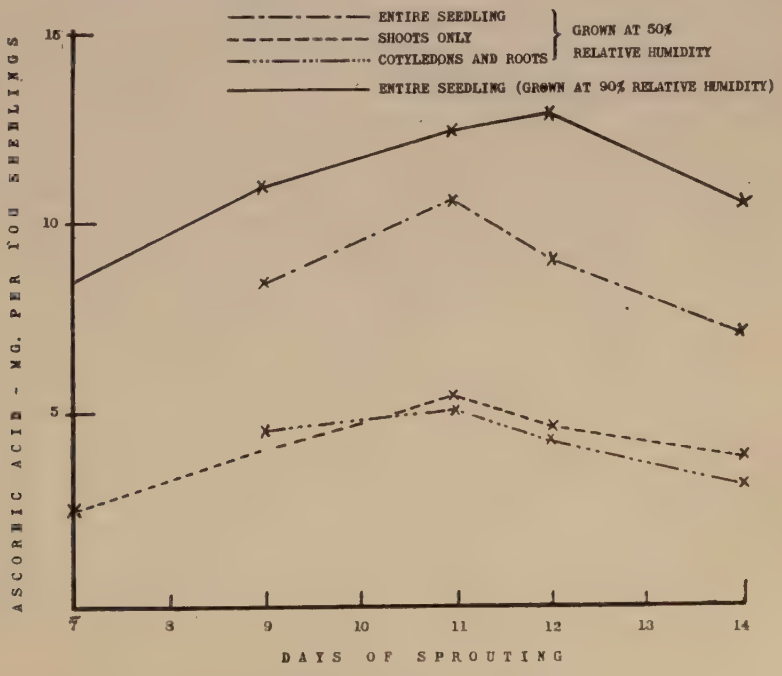


FIGURE 5. Distribution of ascorbic acid in pea seedlings.

The effect of light on sprouting was next studied, the most apparent response being the attractive green colour and retarded growth; the ascorbic acid content per 100 g. of seedlings was unaffected and an apparent increase in the ascorbic acid content due to light was offset by the lower yield. Ascorbic acid synthesis increased only when the light intensity reached a level which permitted photosynthesis. The effect of light can be seen from the data in Table 2, which also shows that above 25° C. there is a decrease in the yield and ascorbic acid content of the green shoots.

TABLE 2.—THE EFFECT OF TEMPERATURE AND LIGHT ON VITAMIN C SYNTHESIS IN 11-DAY PEA SHOOTS

| Treatment | Yield of shoots (gms.) | Ascorbic acid | |
|---|---|---|---|
| | | (mg./100 gms.) | (Mg./400 gms. dry seed) |
| 25° C. in light | 442 | 27.0 | 112 |
| 27° C. in light | 371 | 25.9 | 96 |
| 30° C. in light | 286 | 25.9 | 74 |
| 21° C. in dark | 515 | 22.9 | 118 |
| 26° C. in dark | 431 | 22.5 | 97 |
| 30° C. in dark | 305 | 22.2 | 68 |

The suitability of sprouting bags (Figure 3) for growing green shoots was investigated. Equal amounts of seed were spread on shallow trays and sprouting bags, and it was found that the yield was greater in the latter case, but the ascorbic acid content of the sprouts was almost identical (Table 3). Rot-proofing the bag prevented the roots from penetrating the material thus facilitating the subsequent cleaning of the bag, although the yield in the presence of the copper carbonate was much lower.

TABLE 3.—THE EFFECT OF DIFFERENT SPROUTING DEVICES ON VITAMIN C SYNTHESIS BY FIELD PEA SPROUTS

| Sprouting time (days) | Treatment | Yield of shoots (gm.) | Ascorbic acid content | |
|---|---|---|---|---|
| | | | (mg. per 100 gm.) | (mg. per tray) |
| 11 | Shallow tray | 590 | 26.6 | 157 |
| 11 | Bag, untreated | 730 | 24.5 | 179 |
| 11 | Bag, $CuCO_3$ treated | 496 | 25.2 | 125 |

The results in Table 4 indicate the effect of the thickness of seeding the trays. The yield of shoots increased with increasing amounts of soaked seeds per tray. The thicker the stand the taller the shoots and thus the greater the yield of shoots per 100 g. of soaked seeds. The ascorbic acid content of the shoots increased with the density of the stand but declined when the stand became too thick. The amount of ascorbic acid synthesized per tray was highest where the greatest amount of soaked seed was used, but calculated on a basis of 100 g. of seed, the greatest efficiency was obtained when 1000–1100 g. of peas were spread on one tray.

TABLE 4.—THE EFFECT OF THICKNESS OF SEEDING ON THE YIELD AND ASCORBIC ACID CONTENT OF ELEVEN DAY PEA SPROUTS

| Weight of soaked seeds per tray (gm.) | Yield of shoots | | Ascorbic acid content | | |
|---|---|---|---|---|---|
| | (g. per tray) | (g. per 100 gm. soaked seed) | (mg. per 100 gm.) | (mg. per tray) | (mg. per 100 gm. soaked seed) |
| 814 | 401 | 49 3 | 25.7 | 105 | 12.9 |
| 1014 | 597 | 49.0 | 29.2 | 174 | 17.1 |
| 1144 | 606 | 53.0 | 30.2 | 183 | 15.9 |
| 1414 | 783 | 55.0 | 25.5 | 199 | 14.0 |

The riboflavin content of pea sprouts was determined with the results summarized in Table 5. It is evident that these sprouts would not contribute significant amounts of riboflavin to the daily diet.

TABLE 5.—RIBOFLAVIN CONTENT OF FIELD PEAS BEFORE AND AFTER SPROUTING

| Sample | Riboflavin (micrograms per 100 gm.) |
|---|---|
| Dry seed | 93 |
| 5 day sprouts | 131 |
| 11 day sprouts | 89 |

Seeds and the sprouts were also treated with growth hormones, including the commercial preparations "Parmone", "Stop Drop", and "Auxilin 1". No beneficial effect resulted from their use at high or low concentrations.

After 10 days sprouting, the pea shoots are crisp and green and ready to serve as salads. These are prepared by clipping the shoots close to the seeds, cutting them into pieces 2–3″ long and "tossing" with French dressing. Attractive and nutritious additions include chopped vegetables, such as tomatoes, radish, carrots, lettuce, cabbage, or sprouted soybeans, which have been boiled 20 minutes and chilled.

The residual seeds would not sprout again although they still contained an appreciable amount of storage nutrients and ascorbic acid. This part of the pea sprout represents a complete loss because of the fibrous consistency of the roots. In 10 days sprouting, 400 g. of dry seed produced about 400 g. of green shoots which contained about 120 mg. of ascorbic acid. Therefore, 100 g. of dry seed produced about 30 mg. of ascorbic acid in the form of edible shoots. An average serving as salad, was 40 g. which provided 12 mg. of ascorbic acid.

*2. Navy Beans*

The germination of Navy Beans was retarded by soaking; they had to be sprouted between moist towels or on wet sawdust. The seedlings,

sprouted for 5 or 6 days, contained 11–15 mg. ascorbic acid and 125 micrograms of riboflavin per 100 g. At 12 days a green shoot formed which contained 23–26 mg. ascorbic acid per 100 g. Unlike pea sprouts, the cotyledon of the bean sprouts grew up with the stem to a height of about 4 inches and remained tough and starchy and could not be used in a salad. The cooked sprouts contained only 3–4 mg. of ascorbic acid per 100 g. and therefore their use is not recommended.

*3. Windsor Broad Beans*

The Windsor broad bean (*vicia faba*), sprouted for 4 days, was recommended by Johnson and Young (10) for its high vitamin C content. We found that the beans could be sprouted in tubs, trays or bags. Tubs were preferred as the cleanest sprouts were obtained in high yield and with high vitamin C content (Table 6).

TABLE 6.—THE EFFECT OF DIFFERENT SPROUTING DEVICES ON THE VITAMIN C CONTENT OF WINDSOR BROAD BEAN SPROUTS

| Container | Weight of edible sprouts* | Ascorbic acid content | |
|---|---|---|---|
| | | (mg. per 100 gm.) | (mg. per lot) |
| | gm. | | |
| Tub | 2702 | 25.5 | 690 |
| Open bag | 2639 | 28.2 | 740 |
| Tray | 2511 | 22.8 | 573 |

* Produced in 4 days from 2800 g. soaked seed.

The best results were obtained by soaking the seed for 24 hours and watering twice daily (Table 7).

TABLE 7.—THE EFFECT OF WATERING AND SOAKING ON THE 4-DAY SPROUTING OF WINDSOR BROAD BEAN

| Soaking time (hrs.) | Daily watering | Weight of edible sprouts from 800 gm. soaked seeds | Number of defective seeds |
|---|---|---|---|
| | | gm. | |
| 12 | Once | 785 | 22 |
| 12 | Twice | 871 | 19 |
| 24 | Once | 929 | 13 |
| 24 | Twice | 977 | 6 |

A number of vitamin C determinations were made on several batches of beans sprouted for varying periods of time. The results, presented in Table 8 show that the vitamin C content increased rapidly during the third and fourth day of sprouting. The variability in the results obtained was attributed to changes in temperature during sprouting. Experiments carried out in a constant temperature chamber indicated that the optimum temperature for highest ascorbic acid production was 25° C.

TABLE 8.—SHOWING THE VARIATION IN THE VITAMIN C CONTENT OF SPROUTED WINDSOR BROAD BEANS

| Duration of sprouting (days) | Ascorbic acid—mg. per 100 g.* |
|---|---|
| 2 | 7.1 |
| 3 | 23.1; 14.6; 23.6; 20 4; 17.5 |
| 4 | 25.6; 15.8; 22.0; 16.9 |
| 5 | 28.3; 22.5; 21.2 |
| 7 | 31.2 |

* Each value represents the analysis of a different batch of sprouted beans.

The size of the sprouts was related to their ascorbic acid content and the most suitable stage was found to be when the shoots had broken through the seed coat and attained a length of about 1 inch. Extending the sprouting time did not seem beneficial as micro-organisms spread rapidly and caused spoilage and discoloration of the sprouts. Some bacteria caused proteolytic decomposition which produced a disagreeable odour, while others acted on the carbohydrates of the seed, affecting mainly the appearance and, to a lesser extent, the flavour of the sprouted beans. The danger of contamination by pathogenic bacteria does not seem likely.

Riboflavin determinations indicated that Windsor Broad beans, after 2 days sprouting, contained approximately 1 microgram per gram of fresh material.

The cooked sprouts were utilized in many appetizing dishes. The tough seed coats impaired the acceptability of the beans as a vegetable, but they could be removed by blanching which is a tedious procedure but apparently the only practical one. This was done by dropping the sprouted seeds into boiling water and allowing them to stand for 5 minutes, draining and adding cold water. The seeds were allowed to stand for 3 minutes, when the water was again drained off. When cool, the seed coats had loosened and were easily slipped off. The loss of ascorbic acid in blanching amounted to about 10%. Beans with a disagreeable odour were discarded while those with brown spots were considered edible.

Broad beans were served as vegetable, casserol, stew, or as thick soup. In testing for palatability broad beans were served with tomato sauce to a group of C.W.A.C. personnel. Of 33 answers to a questionnaire twenty-eight considered the beans a tasty addition to the diet, while five disliked them. Twenty-five minutes cooking rendered the beans soft to the point of being mushy but a few remained whole and firm. This gave rise to the comment from a few people that the beans were under-cooked. This opinion was based entirely on texture as only a few minutes boiling is necessary to remove the raw flavour from the beans.

When served as a vegetable with green pepper sauce they were generally very well liked. It was suggested that the beans were too dry and broken up. This was overcome by serving with a more generous portion of sauce and by stirring as little as possible during cooking. Comments on the stew were generally favourable.

Vitamin C analyses on the prepared beans showed a loss of 30–50% of the ascorbic acid originally present in the sprouts (Table 9). Further reduction in the final product was due to dilution by ingredients not containing vitamin C. One helping of thick soup contained about 14 mg. of ascorbic acid.

TABLE 9.—THE VITAMIN C CONTENT OF VEGETABLE DISHES OF SPROUTED WINDSOR BROAD BEANS, AS SERVED FROM AN ARMY KITCHEN

| Vegetable dish | Ascorbic acid—mg./100 gm. | |
|---|---|---|
| | Before cooking | After cooking |
| Mixed with tomato | 20.5 | 12.1 |
| Boiled as vegetable | 25.5 | 11.3 |
| Thick soup | 20.2 | 7.6 |

*4. Vetch*

Vetch seed (*Vicia sativa*) belongs to the same plant family as Windsor broad beans (*Vicia faba*). The sprouts were equally high in vitamin C, containing about 25 mg. per 100 gm. after 4 days sprouting (Table 10). The seed coat of vetch was not as tough as that of Windsor broad beans, and they could be rendered edible by 20 minutes cooking. The taste of vetch sprouts was not as pleasant as that of Windsor broad beans so they were blended with meat or tomato. One serving of sprouted vetch blended with meat contained 5 mg. ascorbic acid, and when blended with tomato, about 14 mg.

TABLE 10.—THE ASCORBIC ACID CONTENT OF VETCH SPROUTS

| Sprouting time (days) | Ascorbic acid (mg. per 100 gm.) | Sprouting time (days) | Ascorbic acid (mg. per 100 gm.) |
|---|---|---|---|
| 2 | 7.3 | 4 | 28.2 |
| 3 | 20.5 | 5 | 37.4 |
| 4 | 23.4 | 6 | 27.5 |

*5. Soybeans*

For the reasons already mentioned the work done on sprouting soybeans was very limited. "Mandarin" soybeans (*Glycine Soya*) were soaked for 8 hours at 22° C. and transferred to the sprouting-tubs. A thin, metal lid was placed on top to exclude light and to maintain a high humidity inside. The sprouts were watered frequently, the water being at about 22° C., and stirred by hand to ensure uniform germination. After each watering the tubs drained slowly and completely thus drawing in fresh air. At the end of the sprouting period (4–5 days) the tubs were emptied into a large water tank where the seed-coats floated-off leaving the sprouted seeds ready for use. The data obtained in these experiments is presented in Table 11. It will be observed that the vitamin C content increased rapidly but failed to reach a high level, and the increase in riboflavin was very slight. The soybeans doubled their weight in sprouting.

TABLE 11.—ANALYSES OF SPROUTED SOYBEANS AT VARIOUS STAGES OF DEVELOPMENT

| Sprouting time (days) | Sample | Cooking time (mins.) | Weight of 250 ml. sprouts (gm.) | Weight of 100 seeds (gm.) | Ascorbic acid (mg. per 100 gm.) | Riboflavin ($\mu$ g. per 100 gm.) |
|---|---|---|---|---|---|---|
| | Dry seed | — | 188.5 | 17.64 | — | — |
| 1 | Raw | — | 165.5 | 38.74 | 1.5 | 76 |
| | Cooked | 60 | — | — | 1.1 | 60 |
| 2 | Raw | — | 175.5 | 40.95 | 3.2 | — |
| | Cooked | 55 | — | — | 1.8 | — |
| 3 | Raw | — | 146.5 | 43.29 | 6.0 | — |
| | Cooked | 60 | — | — | 2.2 | — |
| 4 | Raw | — | 125.5 | 51.17 | 10.4 | 89 |
| | Cooked | 40 | — | — | 1.5 | 40 |
| 5 | Raw | — | 105.0 | 66.16 | 11.4 | — |
| | Cooked | 20 | — | — | 1.7 | — |

It was interesting to note (Table 11) that the cooking time decreased with prolonged sprouting. This was a great advantage as it saved time and fuel. As a result of the shorter cooking time a higher vitamin C retention was hoped for, but this was not found to be the case. Unfortunately the cotyledon required considerable cooking, consequently the rootlets were overcooked. Thirty per cent of the ascorbic acid in the sprout was concentrated in the tender rootlet and this was readily leached into the cooking water.

Sprouted soybeans were served as a vegetable. The 4-day sprouts were about 2 inches long and the beans were clean and in good condition for serving. Before cooking, the beans were washed thoroughly with large quantities of water and as many of the skins as possible were floated off. The beans were first weighed then dropped into sufficient boiling salted water to cover. They were brought to a rolling boil and allowed to cool in this manner for 40 minutes. As a result of boiling, many of the seed coats came to the top and were skimmed off. At the end of the cooking time the beans were drained in a colander (this drainage water should be used in gravy, white sauce or soup stock) and mixed immediately with a tomato sauce.

Of 33 answers to a questionnaire, 13 liked the soybeans as served while 20 disliked them. The general opinion of the latter was that they were not thoroughly cooked. Our experience was that the beans were cooked from the standpoint of flavour but remained tough from the standpoint of texture. In a further experiment the sprouted beans were boiled for 20 minutes, chilled and served as a salad with chopped carrots, lettuce and mayonnaise. The general opinion was that the salad required too much chewing and the flavour was not well liked. Some, however, appreciated the chewiness, so it seemed that a taste for such a salad might be acquired.

## DISCUSSION

The work herein described shows that there are definite limitations in the substitution of sprouts for fresh vegetables. We consider only three types of seed to be of practical importance, viz., field peas, Windsor broad beans, and vetch. Cereal sprouts can only be used in the early stages, i.e. up to 3 days of sprouting. They may then be blended with oatmeal and served as a porridge. Their best feature is their cheapness and abundance, but their nutritional value, so far as ascorbic acid is concerned, is too low to be recommendable.

The attractive appearance and agreeable flavour of pea shoots as a salad met with general approval. Their ascorbic acid content is also high and cooking losses are obviated. The cultivation of pea sprouts, however, takes twice as long as any of the other sprouts. They are the only seed which have to be germinated in thin layers which requires more space and more handling. A tasty and nutritious vegetable soup can be prepared from Windsor broad beans. The ascorbic acid content of the sprouts, originally high, may undergo a considerable loss before serving when cooked too long or not served immediately. The seeds are scarce and expensive. Vetch seeds can be sprouted in a manner similar to that used for Windsor broad beans, but they are not particularly appetizing when served alone. They are palatable when mixed with meat or tomatoes.

The Committee on Food and Nutrition of the American National Research Council (11) has laid down the level for the daily riboflavin requirement at 2.2 to 3.3 mg. for a man weighing 75 kilograms, depending on the degree of physical activity. Considering this as a liberal intake and assuming that 1.8 mg. is sufficient for optimum health, none of the sprouted seeds would contribute a significant amount of riboflavin to the daily diet. The Committee also recommends 75 mg. ascorbic acid daily for the same type of man regardless of his activity. Others (12) believe 30 mg. ascorbic acid is an adequate daily allowance. Recent studies indicate the lower level as being protective against scurvy. With this requirement in mind Windsor broad beans after 5 days' sprouting, common vetch after 4 to 5 days' sprouting and field pea sprouts after 9 to 10 days' sprouting can be recommended as good sources of vitamin C in the diet. One serving of these vegetables will supply one-half the daily requirement of vitamin C. Sprouted soybeans and cereals in the ready-to-serve dish do not contribute appreciable amounts of vitamin C; however, soybeans are valuable for their protein of high biological value and cereals for their vitamin B-complex, so they may be considered a valuable addition to the diet.

Considerable fluctuation was encountered in the vitamin C content of pea sprouts and broad bean sprouts. This was due to variations in the stage of development of the shoot and to changes in external factors during the sprouting period. Reid (13), working with cow pea seedlings, came to the conclusion that the ascorbic acid in the seedlings was partly synthesized from storage carbohydrates and partly through photosynthesis in the young plants. She believed that a simultaneous synthesis and utilization of ascorbic acid took place in the plant and stated that the rate of ascorbic acid breakdown depended on the metabolic rate of the plant. High temperatures increase the metabolic rate, thus explaining the highest vitamin C content in seedlings grown in light at low temperatures.

In our experiment with pea sprouts grown for 11 days, more ascorbic acid per tray was synthesized at 21° C. than at 30° C., while the light of two 100-watt lamps or ultra-violet light had no effect. The failure to increase ascorbic acid synthesis by light was possibly due to insufficient intensity to permit photosynthesis. Ultra-violet light does not stimulate photosynthesis and thus was ineffective in producing vitamin C. Beeskow (14) analyzed Mung beans after 4 days' sprouting and found less ascorbic acid in rapidly growing sprouts than in those which grew more slowly. No similar observation could be made in this work. On the contrary, two lots of beans were brought to the same stage of development in 3 and in 5 days respectively. In both cases the vitamin C content reached 20 to 25 mg. per 100 g.

Cooking methods had a pronounced effect on the vitamin C content of the finished products. Best results were obtained when the sprouts were dropped into a volume of boiling water just sufficient to cover the sprouts and heated rapidly to bring the water immediately back to the boil. The sprouts were cooked in as short a time as possible and served immeddiately and without draining of the cooking water. Nevertheless, 35 to 50% of the ascorbic acid was lost in quantity cookery for army purposes. A further reduction of the vitamin C in the vegetable dish as served was due to dilution with ingredients not containing vitamin C.

## SUMMARY

Methods have been developed for sprouting cereal and legume seeds in bulk, and to utilize the sprouted seeds in the diet in the form of palatable dishes.

Wooden tubs were most useful for sprouting Windsor broad beans, vetch, soybeans and cereals, while shallow screen-bottomed trays were best for peas. An open bag was designed for use in mobile camps and it was found convenient for sprouting all types of seed. Acceptable dishes were prepared from Windsor broad beans, peas, vetch, soybeans and cereals. The beans were best served as a vegetable, thick soup or in stew; the peas in salad; the vetch in meat or tomato mixtures; soybeans in vegetable dishes or in salads; and cereals in porridge or with rice.

It was estimated that one serving (approximately 6 ounces as vegetable, 2 ounces as salad) of Windsor broad beans, vetch and pea sprouts would supply half the daily requirement of vitamin C. The amount of riboflavin is considered to be insignificant.

## ACKNOWLEDGMENTS

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## APPENDIX

### Recipes

*Sprouted Broad Beans—As a Vegetable*

Remove the seed coats from 22 pounds of sprouted Broad beans. Add enough boiling water to almost immerse (ca. 10 quarts), cover tightly with a close fitting lid, bring back to the boil as rapidly as possible and boil 20 minutes. Drain, saving the drainage water for soup stock or sauce, add butter, salt and pepper and serve immediately as a vegetable.

Beans cooked in this manner may be served with many variations of white sauce, e.g., green pepper sauce, cheese sauce, tomato sauce, curry sauce or many other well seasoned sauces.

*Broad Bean Casserole*

20 pounds shelled sprouted broad beans (22 pounds unshelled)
4 quarts canned tomatoes
2 pounds fat pork
8 tablespoons salt
8 onions chopped
4–6 peppers chopped

Mix all ingredients except the beans, heat thoroughly and pour over the shelled beans. Bake in a moderate oven (350° F.) one hour, or until the beans are soft.

*OR*

Simmer gently over low heat three-quarters of an hour or until beans are soft, adding water if necessary.

*Broad Beans in Stew*

In preparing stew for 100 people, 10 to 12 pounds of shelled beans may be substituted for one of the usual vegetables. These are added one-half to three-quarters of an hour before the stew is removed from the heat. Dehydrated vegetables may be used successfully in stew if fresh vegetables are not available.

FIGURE 6. An appetizing and nutritious lunch including sprouted broad bean soup, pea sprout salad and sprouted broad bean casserole.

*Broad Bean Soup*

20 pounds shelled sprouted broad beans (22 pounds unshelled)
6 onions chopped
2 teaspoons celery salt
2 tablespoons chile powder (more may be added if desired)
4 quarts tomatoes
salt
pepper

Remove the seed coats from the beans, add chopped onion and enough boiling water (ca. 10 quarts boiling water to 20 pounds of beans) to almost cover. Boil rapidly one-half hour. Add tomato and seasoning and heat through, stirring to break up the beans. If soup is not thick enough, thicken with flour and butter or flour and water.

*Chile Con Carne with Sprouted Broad Beans*

10 pounds shelled broad beans
6⅔ pounds chopped onions
2⅔ pounds beef suet or other fat
20 pounds coarsely ground beef
5⅓ quarts stewed tomatoes
16 bay leaves
4 quarts water
1½ cups browned flour
13⅓ tablespoons chile powder
salt

Put fat in pan. Fry onions and beans 10 minutes,. Add other ingredients and cook about one hour. Thicken with browned flour. Season to taste.

*Fried or Sauteed Sprouted Broad Beans*

Fry about 20 pounds shelled beans and eight chopped onions in a small amount of fat about 5 minutes. Add a small amount of water and let cook 10 to 15 minutes longer. Serve as a vegetable.

*Pea Sprouts—Salad*

The 10-day shoots were cut as close to the seeds as possible; cut into convenient lengths (2–3 inches); tossed with French dressing and served. The recipe for the salad dressing was:

2⅔ tsp. salt
2 tsp. mustard
2 tsp. paprika
1 tsp. pepper
2 cups vinegar or lemon juice
4 cups olive oil

The bottom of a wide-mouthed pitcher was rubbed with onion. All ingredients were put into the pitcher and beaten until creamy with a Dover egg beater and served at once. This recipe provided enough dressing for 100 salads or more.

Attractive and nutritious additions to this salad were provided by the addition of chopped vegetables such as tomato, radish, carrot, lettuce, cabbage or sprouted soybeans which have been boiled 20 minutes and chilled.

The shoots were also cooked in a minimum amount of water, just enough to prevent burning, and served with butter, salt and pepper as a vegetable like spinach, but this is not recommended as cooking does not improve their quality and destroys a large percentage of the vitamin C.

*Sprouted Wheat Porridge*

1½ cups rolled oats
4½ cups water
4 cups sprouted wheat
½ tablespoon salt

Cook the rolled oats in the salted water 30–45 minutes; at the same time cook the sprouted wheat 10 minutes in water enough to cover, drain and add to the cooked porridge. This provides enough porridge for 13 people.

In preparing a vegetable from sprouted wheat, fry chopped onion and wheat in a small amount of fat 5–10 minutes. (Overcooking will cause the wheat to become very hard.) Add to this, cooked rice and heat thoroughly in the frying pan. This may be used if the supply of rice is short. When used in soup, drop the sprouted wheat into the soup 15 to 20 minutes before removing from the heat. The wheat has a sweet flavour and requires very little cooking. It may be substituted for rice but requires more chewing

## ERRATUM

In the article entitled "A biological method of detecting the presence of fungicides on seeds" by H. W. Mead in *Scientific Agriculture* for March 1945 (Vol. 25, No. 7, pp. 458-461), the illustration on page 459 should bear the following legend.

"Figure 1. Showing the inhibitive effect of two mercury fungicides, carried on wheat kernels, on the growth of *Helminthosporium sativum* on potato dextrose agar. A, Ceresan series; B, Leytosan series. 1, untreated seed; 2 and 3, treated seed."